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THE PRODUCTION OF MECHANICALLY DAMAGED STARCH IN MILLING AS A GOVERNING FACTOR IN THE DIASTATIC ACTIVITY OF FLOUR

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During the last year or two notable contributions to the literature have appeared [1] giving attention to the nature and condition of the starch as the origin of differences in diastatic activity of flours. Sandstedt, Blish, Mecham, and Bode (1937) deduced from the observed course of diastatic activity with time that the higher activity found with commercial (American) flours than with laboratory-milled was due to the fact that the former contained larger proportions of the more susceptible starch fractions, in other words greater amounts of ruptured starch granules. Blish, Sandstedt, and Kneen (1938) point out that autolytic diastasis in flour is very rapid at first but after an hour or more diminishes to a very low rate, a fact attributable to exhaustion of the available starch substrate, not to exhaustion of active enzyme. They further point out that the small amount of the starch normally converted is substantially increased through additional grinding of the flour as in a ball mill and that it is apparent from their data on autolysis that a portion of the "susceptible" starch (susceptible to beta-amylase attack) in flour is a result of the mechanical rupture of raw starch during milling. They add that it seems likely, however, that at least a significant amount occurs naturally in the form of dextrins or closely related substance.

L. H. Pulkki (1938) observed marked increases in maltose figure and gassing power in samples of middlings ground to pass increasingly fine silks. He attributed this to the increase in the proportion of mechanically damaged starch granules which could be stained with congo red.[2]

[1] Since the preparation of this paper for publication, O. E. Stamberg and C. H. Bailey (1939) have reported that although alpha-amylase (made from germinated wheat) can hydrolyse substantial proportions of raw wheat starch, beta-amylase (made from normal wheat) is able to hydrolyse only small amounts. They show however that when the same wheat starch had been severely mechanically damaged it was hydrolysed as easily as soluble starch paste by both alpha- and beta-amylase.

[2] Actually C. L. Alsberg and E. P. Griffing (1925) had covered substantially the same ground in this respect except that they dealt with the amount of cold water extract obtainable instead of maltose figure. They observed (qualitatively) that the proportion of starch granules stainable with congo red increased markedly as a sample of flour was ground to pass progressively finer silks. They in turn were directed to the use of congo red solution by an earlier finding of H. Huss (1922).

He considers that the starch granules of wheat have a separate thin protective surrounding layer or film, which unlike the inside material of the granule is not easily attacked by diastase nor stainable with congo red but can easily be removed by mechanical means. This in the present writer's view comes very near to the truth but does not quite accord in a few important particulars with some observations to be described. This and certain other aspects of Pulkki's paper will be further considered in appropriate places in the present paper. It might however be added here that Pulkki thinks that "if some entirely different method of grinding (other than rolls) were used, it would be at least theoretically possible that a high degree of fineness could be reached without affecting very great changes in amylolytic activity of the material."

The conclusions and views cited above had in fact formed a material part of a position reached by the writer some years ago as set out in Reports of the Research Association of British Flour-Millers [3] and he is naturally gratified by the support now appearing for his observations and deductions. In view of the extent of the interest now taken in this field he feels it may be of interest and service to other workers to put forward some of the further conclusions arrived at during the work in question.

It might be illuminating to describe briefly how the matter came to receive attention in the first place. A laboratory mill had been in use in the St. Albans laboratories for many years and in 1933 a second new mill was obtained and put into additional operation. Comparisons of flours made from similar wheat samples on the two mills, after careful attention to equivalent operation in every obvious particular, showed marked differences in maltose figure and gassing power. Systematic tracing of the factor responsible made it clear that the difference in reduction-roll surface was responsible. The rolls of the new mill had a smooth, polished surface; the old were rather rough and pitted.

Microscopical Appearance of Starch Granules Damaged during Milling

The microscope was then used to ascertain the nature of the change in the flour leading to raised maltose figure as a result of the use of rather rough reduction-roll surfaces as against smooth under quite similar conditions. In the first instance it was not known what to expect beyond the probability that one flour would show some form of starch damage absent from the other. One expected (*a priori*) to

[3] Confidential Reports of the Research Association of British Flour-Millers, Nos. 28, 30, and 31, issued in February, September, and October, 1934.

see some of the granules cracked or split, probably radially on the lines of the preparation shown in Plate 1.[4]

This type of damage, however, does not occur in flours freshly roller-milled, and it may be said at once that the type of damage which actually is found associated with an increase in maltose figure, due to one form of rolling as opposed to another, is less obvious, though it is necessarily seen to varying extents whenever any flour is examined microscopically.

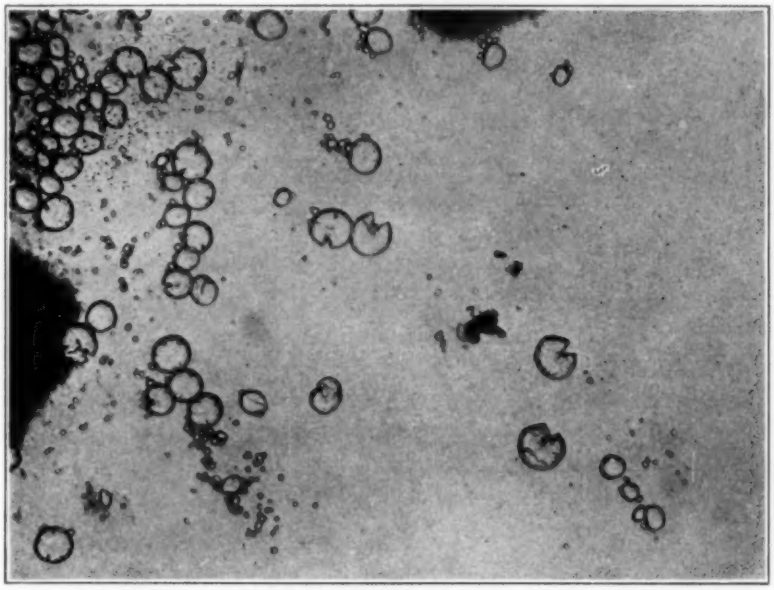


PLATE 1

Granules of wheat starch cracked radially through hand-manipulation on the slide.

(All magnifications are approximately × 120 diameters)

The damage is nevertheless highly characteristic. A flour which has been fairly severely milled (using the word "severely" in the sense of conditions which make for production of high maltose figure) is shown in Plate 2A.

The starch granules in question which have sustained damage are indicated by a star on the photograph. They do not look cracked but have a curious flat appearance and thin faint outline. Compare for example *a* in Plate 2A, with the sound granules marked *b* or *c* (the latter in the upper left-hand corner). Their attenuated appearance in contrast to the boldness of sound granules suggested the term "ghosts,"

[4] The magnification in each photograph is approximately 120 diameters.

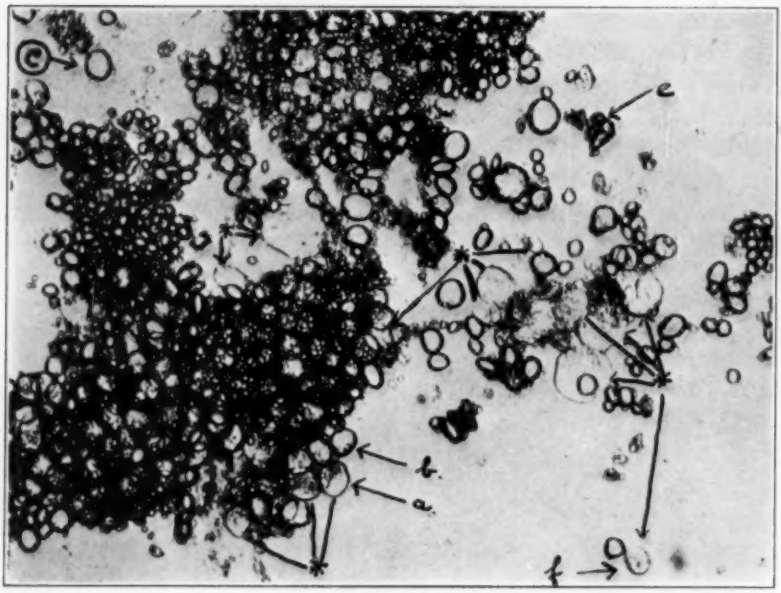


PLATE 2A

Wheat flour showing "ghosts."

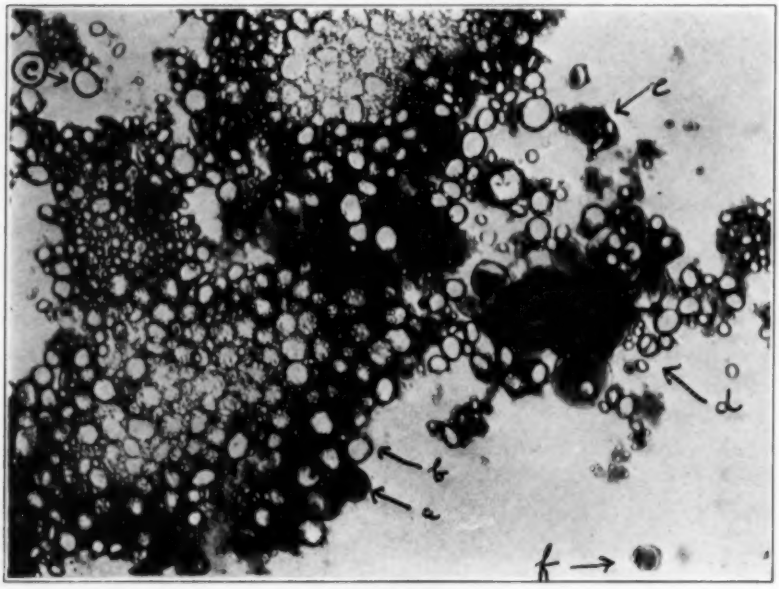


PLATE 2B

Same preparation as 2A afterwards stained with congo red.

which was afterwards found not inconvenient in referring to this particular type of starch damage.

"Ghosts" are more easily detected in flour through suitable differential staining, a phenomenon which is also of significance in regard to the part they play in diastasis. Dilute aqueous congo red solution is excellent for the purpose (0.1% to about 0.35% is a suitable concentration). It does not stain sound starch whilst the "ghosts" all stain uniformly an orange-pink. In flour preparations it stains gluten a rather vivid brown. Care is sometimes required (especially with low-grade flours) to avoid confusion with pieces of endosperm cell-wall tissue, which stain a vivid pink.

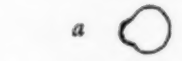
Plate 2B shows the same preparation as 2A after staining [5] through irrigation with 0.35% congo red solution. It is interesting to compare now the appearances of the granules referred to above and also to note how the colony of "ghosts" at *d* in Plate 2B is brought into prominence. Incidentally the gluten, which, in the form of its delicate meshwork permeating the large clumps of starch granules, is only indistinctly apparent in Plate 2A, is dyed intensely by the stain and its extent becomes sharply defined in the photo. This is well shown, for example, by the very small endosperm particle *e* in Plates 2A and B, and also in patches at *d*. With the very large particles, however, the stain is taken up eagerly by the gluten of the outer margin, the interior escaping staining as is well seen in Plate 2B.

The results of staining by irrigation are easily deceptive in many ways in that uniform distribution of the stain over the field cannot be brought about. In Plate 2B, for example, the stain streamed in from the lower edge of the photo with the consequence that the upper edges of particles tended rather to escape staining relative to the lower. The streaming also produces a certain mechanical derangement of the objects under view. The relatively large mass of which *a* and *b* are outlying granules has thus been shifted upwards, approaching the upper mass of roughly equal size. The gulf or inner sea initially separating the two has thus been contracted and partly forced out towards *e*. The currents of plain water so formed further upset the distribution of the dyestuff. The upshot is that many of the particles seem from the photo to show an irregularity or non-uniformity of staining which is apparent rather than real. In the great bulk of the cases to be described, uniformity of staining was obtained by making up the preparation at the start with the stain solution.

A few very important features of this staining must now be mentioned. It is uniform: starch granules either stain alike or not at all.

[5] The effect is difficult to register photographically with the vividness presented to the eye. The artifice was found helpful of using green light in conjunction with ortho-chromatic plates.

Every stained granule has the same tint, and each granule is uniformly tinted through its stained part. It is possible very occasionally to see a granule which is partly sound and partly "ghost," shaped thus:



the part *a* having the "solid," strongly outlined appearance of sound starch and *b* the customary faint, enlarged outline of the "ghost." In such a case *b* stains uniformly, *a* remains unstained. Apart from such rare cases the "ghosts" are invaded by the stain from all around the circumference equally. This may be seen if, instead of making up the slide initially with dye solution, an attenuated preparation with water only is made and irrigated with the congo red solution under the microscope. The zone of stained material in any given "ghost" increases in width uniformly from the periphery inwards as the colour encroaches evenly until the last unstained point at the center has disappeared leaving the "ghost" uniformly tinted. This process is exceedingly rapid with congo red solution; it happens so quickly that it is not easily followed. It is slower and much more easily followed with sufficiently dilute iodine solution.

Iodine (in an aqueous solution of potassium iodide) of more than a certain concentration stains all starch granules, sound or damaged, indiscriminately a very deep purplish blue. Such an effect is shown in Plate 3A, the solution used containing 0.3% iodine. On irrigating a flour-water slide with such an iodine solution, however, it is possible to see the "ghosts" take up the colour much more quickly, and at first more deeply, than the sound granules. It was then found that if the iodine solution used for making up a slide is sufficiently dilute the "ghosts" will stain intensely blue, whereas the sound granules will never stain more than a very faint mauve. A solution containing 0.02–0.03 g. iodine and 0.07 g. potassium iodide in 100 g. of water gives in this way an exceedingly sharp differentiation between "ghosts" and normal granules. The effect is shown on Plate 3B. Apart from intensity, the colours of the normal granules and the "ghosts" are distinct, the former being purple and the latter pure blue.

A further important feature of the staining phenomena is that the process (whether with iodine or with congo red) can be reversed. If the stained slide is irrigated with pure water the stain disappears from the "ghosts," from the outer margins evenly first and lastly from the center. The process can be repeated many times, fresh dye being introduced and then removed. After several repetitions, however, the intensity of the stain taken up by the "ghosts" becomes much less.

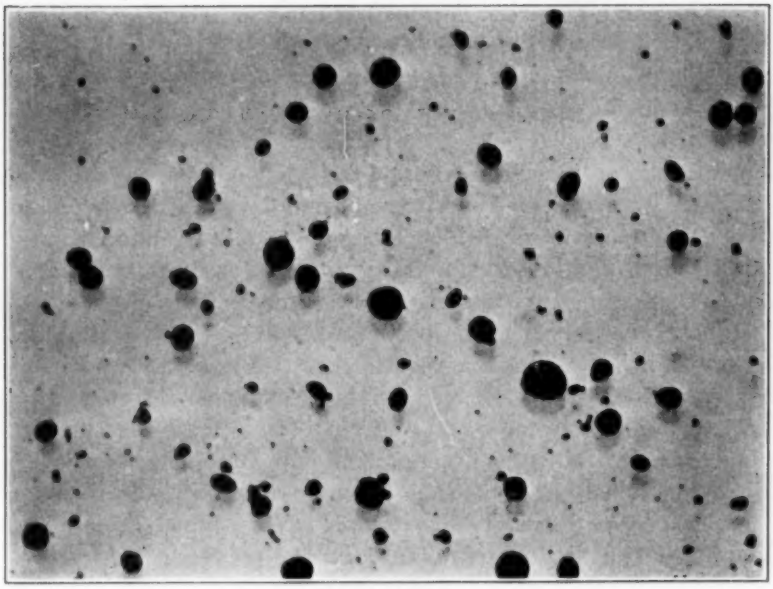


PLATE 3A

Wheat starch, partly sound and partly "ghosts," stained with 0.3% iodine solution.

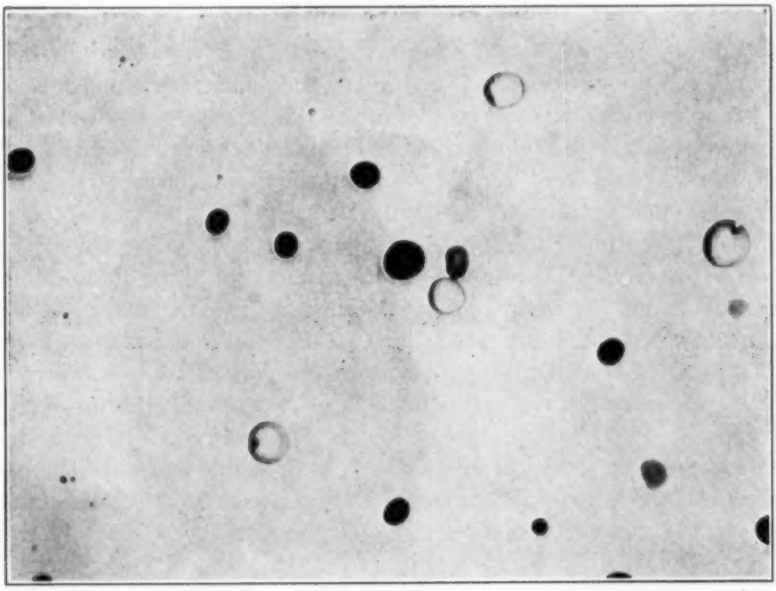


PLATE 3B

Wheat starch, partly sound and partly "ghosts," also with a radially cracked granule, stained with 0.02% iodine solution.

Some Different Forms of Mechanical Damage to Wheat Starch Granules

The larger granules of wheat starch may be regarded as playing the predominant part in the characteristic phenomenon under discussion. Usually reported as of 30 to 35 μ in diameter, they are lenticular in shape but ordinarily are seen lying flat, with a circular outline. Mechanically they are very delicate structures and if gentle pressure is applied to the cover slip whilst the granules are examined under the microscope it may be seen that they are very easily flattened with, of course, a resulting increase in the diameter of their circular outline. They have marked elastic properties. If with a microscope slide of a mixture of flour or starch and water the cover slip is gently pressed, any given granule affected by the pressure expands perfectly uniformly in area as viewed from above, and on release of the pressure returns immediately to its original shape. If now the cover slip is pressed rather more firmly in a "springy" fashion and released, pressed again and released, the process being repeated several times (preferably, it is thought, with a slight interval between the releasing and re-pressing) the granules crack or split in a characteristic radial fashion, the damage produced being shown in Plate 1.

If, however, a fresh slide is made and the cover slip handled rather differently, an entirely different type of damage is produced. In this process a much heavier pressure is applied gradually and maintained for a little time so that the granules are not released but kept just beyond what might be called the limit of their elastic deformation. On now releasing the pressure the granules do not recover as in the first case. They are permanently flattened and have become in fact "ghosts." As a very rough estimate they now have 50% greater diameter than at first.

If the treatment is rough the "ghosts" produced are extensively cracked or fissured finely in a radial fashion for some little distance inwards from the periphery; with very rough treatment they can eventually be broken up. In fact, it requires some practice to produce the ghosts "artificially" in this way in the perfect condition in which they are nearly always found in flour as a result of ordinary milling processes. Perfection of condition is here spoken of as meaning freedom from all cracks or fissures and easily visible flaws, the "ghosts" as mentioned previously having characteristically the same outline appearance viewed from above as the sound granules. Plate 4A shows "ghosts" made by hand manipulation in the way described, the granules in question having all been sound when the slide was first made. The most nearly perfect examples are denoted by an X marked on the photograph.

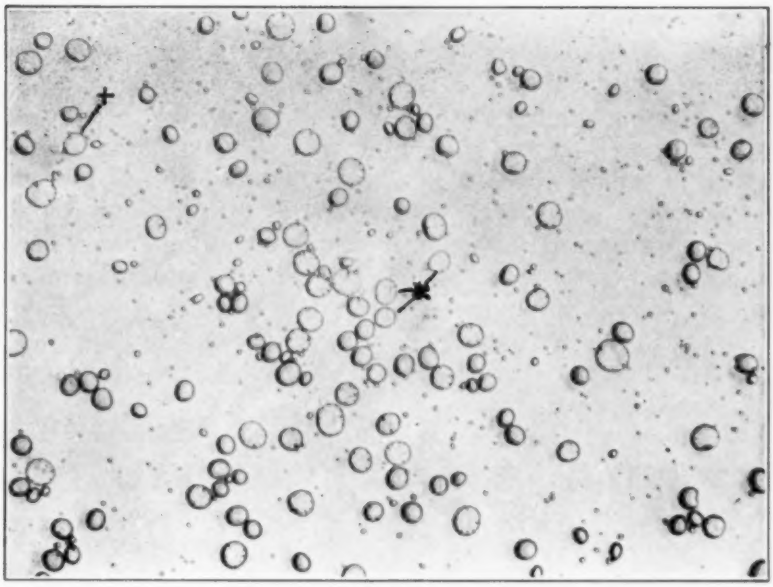


PLATE 4A

Wheat starch containing some "ghosts" made by hand-manipulation on the slide, unstained.

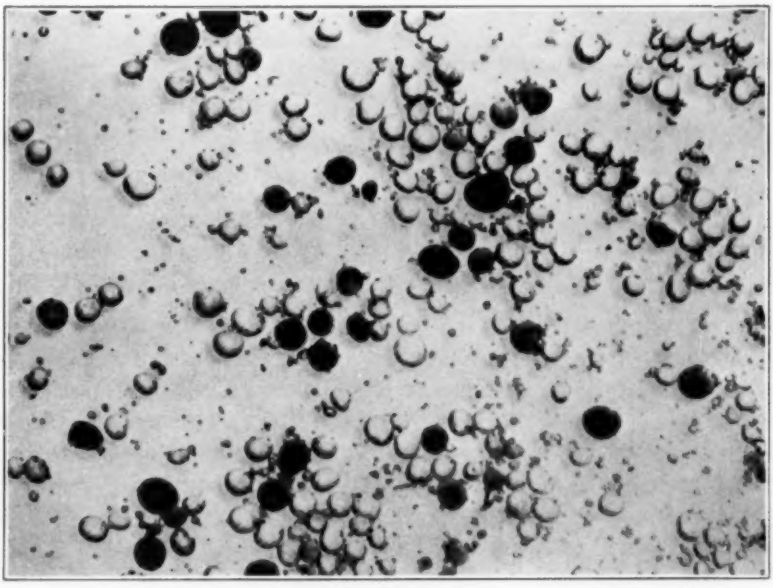


PLATE 4B

Wheat starch containing some "ghosts" made by hand-manipulation on the slide, stained with 0.03% iodine solution.

In every way, as far as can be observed under the microscope, the "ghosts" so produced behave the same as those found in the first place in flours. They stain in exactly the same way with congo red or the 0.03% iodine solution. Plate 4B shows a preparation in which some of the granules have been made into "ghosts" by hand manipulation: all such are stained with 0.03% iodine. On the other hand, the radially cracked "solid" granules do not stain except perhaps for a very slight margin on the edges of the larger fissures when these are V-shaped: they behave like the sound granules with respect to staining reagents. Plate 4C gives a very clear view of a few granules in 0.02% iodine solution. One is a "ghost" made by hand manipulation and has stained at once, in contrast to the others, two of which are sound and two radially cracked.

It is thought, however, that on long standing the radially cracked granules slowly take up the stain, eventually becoming uniformly

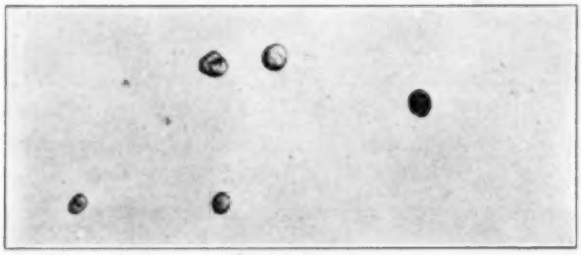


PLATE 4C

A small group of wheat starch granules in 0.02% iodine solution.

stained throughout, whereas the perfectly sound granules remain unaffected in the presence of the stain.

We thus have two characteristic, different forms of damage to which the starch granule may be subject of which one, the "ghost" type, is specifically a result of flour-milling processes generally.

It should be emphasized that the fissuring of the starch granules produced during *germination* of the grain is a quite distinct type of damage. As is well known the granules are "pitted" under these conditions. Even at an advanced stage, however, when badly eroded starch is concerned, the granules so attacked do not stain with congo red,[6] though it was found that the 0.03% iodine solution will stain them, not however a pure blue as in the case of the "ghosts," but a deep purplish colour.[7]

[6] This phenomenon is one of the pointers indicating that in ordinary damage of wheat starch it cannot be a question of peeling off a protective skin as suggested by Palkki.

[7] It is outside the scope of this paper to discuss non-mechanical means of damaging starch granules, *viz.* by heat in presence of water or by means of chemical reagents. It may however be pointed out that granules heated in water to just above the gelatinisation temperature appear, stain, and behave like "ghosts" produced mechanically as described above.

One Type of Damage in Milling Practice

The point the writer wishes to make now is that of these various types of damage it is only the "ghost" type that is produced during ordinary flour-milling operations. It will presently be shown that a number of very distinct mechanical factors in flour-milling reduction processes produce damaged starch, but always the granules affected are in the form of "ghosts." Moreover they are produced exceedingly easily and it is not surprising that all commercial breadmaking flours contain appreciable though varying amounts. In fact, unless the grain is well soaked to soften it, it is impossible to cut a wheat grain and remove particles of endosperm from the cut surface with a knife, however gently, without damaging much of the starch. Again the striking thing is that starch so damaged is in the form of "ghosts." By shearing (as in scraping) or by heavy pressure (as on the microscope slide) the one characteristic type of damage is produced—a point which is of practical importance. Another point no less important is a property of the "ghosts" when found under certain circumstances of occurring in *aggregates* which are always in the form of flattened flakes.

A small flake or sheet of "ghosts" may be seen for example attached to the "southwest" side of the flour particle near the center of Plates 5A and 5B. Often the flakes are much more extensive than this and they may be found in particles much larger than those of flour, *e.g.* dunst.

On gently rubbing the cover slip over the slide the colonies of "ghosts," whether alone or attached to a starch-gluten aggregate, are clearly seen to have relatively considerable cohesion. They hang together very well whereas the sound starch, once it is gently freed from the gluten meshwork, comes teeming out in discrete granules. It will be shown later how this property is of practical importance.

The Relation between Number of Ghosts and Diastatic Activity

Plates 5A and 5B show a picture of a low-grade flour (*L*-roll flour) which had a maltose figure of 3.5%, having come from a grist rather abnormally rich in diastase. As distinct from higher-grade flours, no very large aggregates are to be seen, the starch granules being more or less scattered, and amongst the larger granules the "ghosts" outnumber the sound granules.

On standing overnight a remarkable change was seen in the slide of the *L*-roll flour. Hardly any "ghosts" could be seen, the starch being present as sound granules, which did not appear in any way altered. Fresh irrigation with congo red solution produced no further staining at any point in the slide. It is not possible to show the

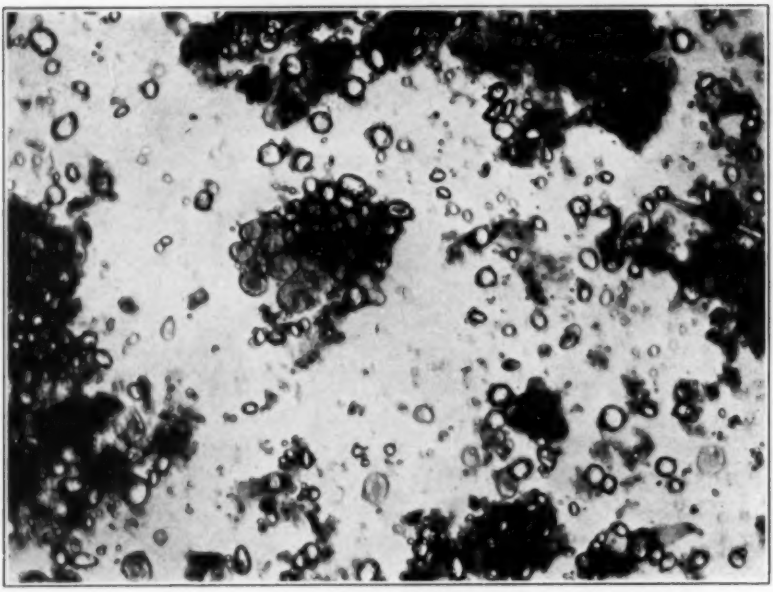


PLATE 5A

An *L*-roll flour (maltose figure 3.53%) (stained with congo red).

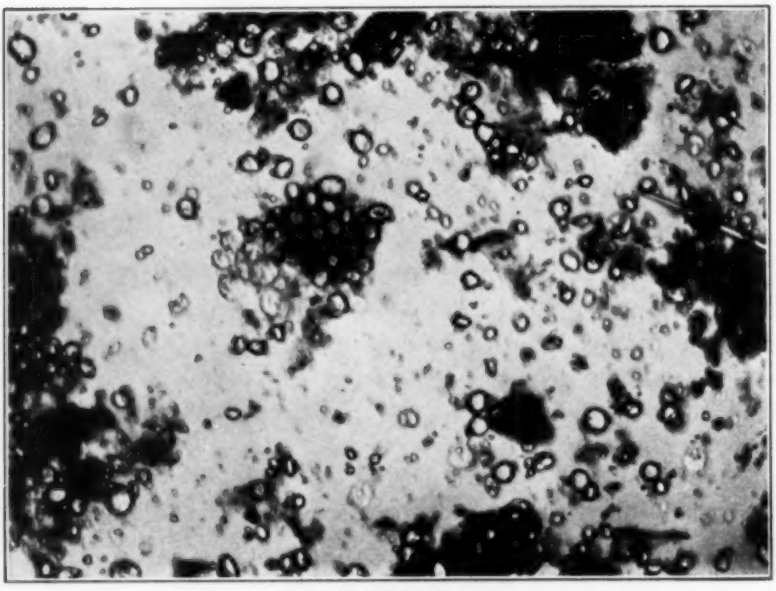


PLATE 5B

The same preparation as 5A after standing untouched a few hours.

phenomenon here in the marked degree described but nevertheless comparison of Plate 5B with 5A is very informative. Plate 5B was the same subject taken after standing a few hours, 5A, of course, having been taken immediately the slide was made up. It will be seen, for example, how the big patch of "ghosts" near the center has almost faded away, whereas the initially sound granules are in general unaffected. By contrast a stained slide of laboratory-milled low-grade flour from Plate wheat (of low diastatic activity) which had maltose figure 1.0%, showed relative permanence in what "ghosts" it contained; even after standing two or three days the "ghosts" were visible, distinctly stained. If such a flour, or even commercial wheat starch, is made up on the slide with a solution containing highly diastatic malt extract in addition to the congo red stain, any "ghosts" that are present practically disappear in less than ten minutes at room temperature, the sound granules appearing unaffected.

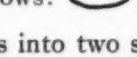
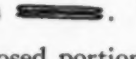
In general with flours differently milled from the same wheat the order of abundance of "ghosts" is in the same direction as the maltose figures. Moreover, the range of maltose figure which can be covered in the case of samples of flour milled from a given sample of conditioned wheat is very great. Thus in the case of No. 2 Manitoba wheat, flour of maltose figure 1.0% was obtained with a very gradual laboratory milling system (gentle reductions, low releases, and correspondingly numerous passages through the rolls) in which highly smooth reduction rolls were used. By the use of a similar system with rather rough, pitted reduction rolls on the same sample of conditioned wheat, a similar length of flour of maltose figure 1.9% was made. Using the rougher rolls with fairly heavy pressures and fairly high releases and correspondingly fewer reductions, the maltose figure was 2.2%. Moreover, if any of the finished flours were taken and put a few consecutive times through closely engaged smooth rolls the maltose figure could without difficulty be raised to something of the order of 5%, an observation in line with those of several other workers.

Damaged Granules in Diastasis

The facts so far given led, as has since been substantially recognized by other workers, to the position that only the "ghosts" could be regarded as undergoing diastatic conversion in ordinary flour doughs or suspensions.

It is outside the scope of this paper to consider closely the structure of the starch granule and the various conceptions advanced by many workers. It will suffice to make the simple statement that the soluble "inner starch" is ordinarily protected by an envelope or skin of

insoluble "outer starch" surrounding the granule. The former alone is fermentable by the diastatic agent present in ordinary sound non-germinated wheat flour. This agent cannot act until the outer resistant skin of the granule is broken, either by mechanical means or through heat gelatinisation. Therefore the "ghosts" alone are the source of sugar formation in ordinary dough, and the greater their number the higher is the maltose figure.

The microscopic observations recorded suggest that the characteristic mode of damage resulting in "ghosts" consists of a completely peripheral rupture or cleavage between upper and lower dish-shaped skins of the initially sound lenticular granules. The crude diagrammatic illustration is as follows:  becomes  . The material of the granule divides into two superimposed portions. That the effect is unique in the completeness with which the interior of the granule is opened up to the entry of water and solutions is shown by the staining behavior.[8]

Where the staining reagent can enter freely the diastatic enzymes can penetrate and the products of their action can be removed outwards through diffusion. The observations described on starch to which malt extract has been added, and on a flour preparation which was rich in diastatic enzymes, leave no doubt as to this conclusion. The "ghosts," and only the "ghosts," were seen to disappear entirely. On the other hand, with a flour which was relatively poor in diastatic activity the "ghosts" persisted over a long period.

It will be realized that flours are able to behave in the accustomed way during fermentation only because of the fortuitous circumstance that they all contain some mechanically damaged starch, *i.e.* "ghosts." Flours, however, vary greatly in their content of "ghosts" and it has not perhaps been generally realized how fundamentally this consideration affects the whole question of the fermentation and gassing power of flour. Content of damaged starch and amount of enzymes are factors which are entirely independent but which together determine the diastatic activity of a flour as ordinarily observed.

Diastatic Activity and Particle Size

From his observations the writer thinks that any factor other than the two discussed above, such as the question sometimes put forward

[8] Congo red is taken up by the inner starch but not by the skin. As soon as the latter is ruptured in the manner suggested, the stain enters at once uniformly from all points round the periphery. That the entry, and exit too, of the stain and of water is free, and that the inner starch is readily dissolved out of the "ghost" are shown by the behaviour described under repeated irrigation and re-staining: in the end the intensity with which the stain is taken up decreases, suggesting that much of the inner starch, which alone stains, has been removed. The behaviour with iodine is even more significant because it is well known that different members of the starch family show different colours with iodine. The soluble, inner starch gives a pure blue; the skin or envelope, in the iodine concentrations used by us, colours mauve.

of mechanical accessibility of the starch granules in bulk (whether or not they are freed from the gluten meshwork), is practically insignificant in effect beside the two main factors here postulated. Certainly it will become sufficiently clear, from observations to be described, that granularity, or particle size, of flour and milling stocks is not for practical purposes a factor determining diastatic activity. The maltose figure obtained through autolysing a suspension, even of coarse endosperm particles like coarse semolina, for a given period may be taken as substantially equal to that which would be obtained were it possible to reduce the particles to fine flour without further starch damage, and then to autolyse this flour under similar conditions.

Distribution of Diastatic Enzymes in the Endosperm

This statement logically has an important consequence; or, rather, one might say that it depends for its validity on a fact which it is important to establish—namely, that the diastatic agent responsible for the attack on the "ghosts" in ordinary non-germinated material is uniformly distributed through the endosperm. If so of course it is easy to understand that particle size *per se* does not matter. It will not be a question of an enzyme (*i.e.*, a colloidal substance) having to diffuse into the interior of a conglomerate particle in suspension. The enzyme will be there already, uniformly in all parts of the conglomerate, merely waiting for the comparatively easy and rapid penetration of moisture, only, to come into activity.

That this is so is established beyond reasonable doubt through the following experiments.

A sample of well purified high-grade semolina was obtained. It was so prepared that it may be regarded as representing predominantly the inner portion of the endosperm, contaminated to the minimum possible extent with particles of bran coat, germ, and the endosperm from nearest the bran. Samples of it were reduced to flour in the laboratory mill in three different ways.

A mixture of 75% of this high-grade semolina was then prepared with 25% of a fifth-break (last-break) semolina prepared from a mixture of wheats. This last-break semolina was of course heavily contaminated with bran snips, pieces of germ, and generally with particles of endosperm scraped off the bran coats. The 75%–25% mixture was then reduced in the three ways comparably with the work done on the high-grade stock. The respective flours were compared. The details are given in Table I. It should be explained that the semolina (over 60 g.g.) was reduced several successive times until it afforded (1) rejected semolina offal or semolina "tails," (2) dunst (through 60 g.g.

over 10 s.), and (3) semolina flour (through 10 s.). The dunst was then taken and rolled repeatedly until a satisfactory finish was obtained, the products being "dunst offal" and dunst flour. All weights recorded are on the basis of 1.5 Kg. of semolina fed to the rolls.

TABLE I

THE MALTOSE FIGURES AND ASH CONTENTS OF FLOURS MADE BY REDUCING, UNDER VARIOUS CONDITIONS, SEMOLINAS AND DUNSTS

1. Of high purity (representing chiefly the inner portions of the endosperm).
2. Mixed with 25% of offally stock (containing pieces of bran, germ and endosperm from near the bran coat).

Test No.	Conditions of reductions	Semolina reduction				Dunst reduction			
		Wt. of semo. flour	No. of rollings of semo. required	Ash content of semo. flour	Maltose fig. of semo. flour	Wt. of dunst flour	No. of rollings of dunst required	Ash content of dunst flour	Maltose fig. of dunst flour
	Using high grade semolina								
		g.		%	%				
1 A	Fast running rolls and heavy pressures	416	4	0.412	2.1	985	6	0.342	1.9
1 C	Fast running and light pressures	363	7	0.436	1.9	1048	10	0.324	1.7
1 F	Slow running and light pressures	345	8	0.448	1.8	1030	9	0.348	1.8
	Using 75% high grade + 25% last break semolina								
2 A	As for 1 A	376	4	0.560	2.0	949	6	0.404	1.9
2 C	As for 1 C	328	7	0.628	2.0	956	10	0.414	1.8
2 F	As for 1 F	353	8	0.580	1.7	897	9	0.412	1.7

The flours of the second series were reduced under rolling conditions comparable with those of the first, and it can be taken that respective pairs of flours (*e.g.* 1A and 2A, 1C and 2C, and so on) have similar contents of damaged starch. Those of the second series, however, are considerably richer in endosperm material from near the bran and more contaminated with finely comminuted bran and germ—to an extent which can be judged from the markedly higher ash contents of the second series. Despite this, *the maltose figures of the flours of the second series are substantially no different, respectively, from those of the first.* That is to say, the introduction of bran, germ, and a different portion of the endosperm in significant amounts has made no difference to the extent of sugar production in one hour from a given proportion of damaged starch. Had even a small amount of diastase been added to any of these flour suspensions we know well that the sugar production in one hour would have been appreciably raised.

The writer submits the conclusion therefore that the diastatic agent responsible for sugar production in flour from normal grain is uniformly distributed throughout the endosperm.

Maltose Figure as an Index of Content of "Ghosts"

On the foregoing basis of course, *with a given wheat sample* and over the range of maltose figures ordinarily encountered in technical practice, the maltose figure may be taken as a strict measure of the quantity of "ghosts" made through any particular milling process. This is very convenient [9] and has in the writer's opinion amply proved itself in extensive investigations he and his colleagues have made on the interplay of milling factors. In the present work the comparative maltose figures given will be taken as measuring comparative amounts of starch damage.

At the risk of repetition perhaps it may be mentioned here that in the case of large particles of endosperm, as in the coarse semolina fresh from the break system, the very low maltose figures usually observed are not low because of the large size of the particles. They are low because the proportion of "ghosts" is very small.

Damaged Starch in Primary Stocks and in Grinds from Reduction Rolls

One must sharply distinguish between stocks straight from the breaks (what may be called virgin or primary material) and stock which has, once at least, passed through reduction rolls, however gently set.

TABLE II

THE TREND OF MALTOSE FIGURE WITH PARTICLE SIZE IN PRIMARY STOCKS (UNREDUCED STOCKS STRAIGHT FROM THE BREAK SYSTEM)

	Through 24 g.g., over 1 silk	Through 1 s., over 5	Through 5 s., over 10 s.	Flour (through 10 s.)
Corresponding range in sieve apertures (*mm.*).....	0.86–0.39	0.39–0.27	0.27–0.135	—
Maltose figures: set				
No. 1.....	0.45%	0.41	0.62	1.39
No. 2.....	0.39	0.39	0.54	1.38

In the case of primary stocks of different particle size there is little or no tendency for maltose figure to rise as the particles become finer, at any rate down to about No. 10 silk. This is exemplified by the figures in Table II obtained on two sets of stocks sifted from two lots of purified commercial semolina made from the same wheat.

[9] It saves the need for direct estimation of the proportion of "ghosts" which, as will be shown in a later communication, if attempted on washed-out starch (as by Pulkki) may give rise to misleading conclusions.

When coarse primary particles are passed through reduction rolls once, even very gently, the particles of different sizes show a characteristically different series of maltose figures. For example the mixture of the above-mentioned primary stock through 24 g.g. over No. 1 silk, in sets No. 1 and No. 2, was reduced once very gently between smooth rolls. The grind was sifted and the maltose figures of the collected respective fractions are shown in Table III.

TABLE III

THE TREND OF MALTOSE FIGURE WITH PARTICLE SIZE IN *A*-GRIND IN THE CASE OF A GENTLE REDUCTION OF SEMOLINA BETWEEN SMOOTH ROLLS

Through 24 g.g., over 5s.	Through 5s., over 10s.	Flour (through 10 s.)
1.28%	0.61%	1.32%

Quite generally the maltose relationships of primary stocks of different particle sizes on the one hand and of stocks produced by the partial reduction of coarse primary particles on the other may be represented as follows:

Damage Due to " Internal " and " Surface " Factors

In seeking to understand the important general relationship described above it is necessary to distinguish between two factors at work on any given particle of stock in any reduction process. Each factor produces the same form of starch damage—"ghosts"—but in different ways. They may conveniently be called the "internal factor" and the "surface factor." To remove ambiguity let us state that the surface factor involves consideration *both* of surface of the particle and of surface of the roll. It is the shearing or scraping effect on the surface of the particle by the surface of the roll that produces a certain proportion of "ghosts" in the flour made by the scraping, the proportion produced being characteristic of the type of roll surface.

If we have "virgin" particles of semolina and subject these to some process involving gentle attrition with a minimum of pressure, such

as the gentle attrition which takes place in spouts and elevators, the whole of the flour produced is from the surface of the particles. In such a case the maltose figure of the flour produced is fairly similar to, or very slightly higher than, that produced by smooth rolls working very gently on the same semolina.

Thus the flours of Table II are really "attrition flours," the maltose figures being of the same order as that of the flour made by smooth-roll reduction of the same parent stock (coarse semolina) in Table III.

When uniformly coarse particles of endosperm are rolled it must be assumed that all sustain first a surface effect, some flour and perhaps some dunst being removed from their surfaces. It may be assumed too, though it is not highly material to the present argument, that some of the coarse particles will then offer little resistance to the approach of the nip, but will crumple up on light impact into flour and much stock of intermediate sizes, especially dunst. Such products will have "come off lightest."

The Internal Factor

The important point is that others of the coarse particles offer more resistance and are not disintegrated but are merely more or less flattened. Such particles have resisted for the moment material reduction in size, but they have undergone severe strain, the strain being the greater the greater their size. The results of the strain are shown in two ways:

(1) A mechanical weakening of the structure of the particle so that subsequent disintegration is easier;
(2) The conversion into "ghosts" of the whole of the starch granules within more or less well-defined zones, or rather probably planes, within the particle.

This latter is the *internal factor* in the production of starch damage during the first rolling of semolina. It takes place completely within the particle, away from any possible direct influence of roll surface, as a result of the internal shearing strain due to pressure in the organized structure of the particle. The production of "ghosts" in this way is greater the greater the work done on the particle, *i.e.* the larger and the more resistant the particle and the heavier the pressure used. Naturally therefore the resultant increase in maltose figure of the coarser stuff is found to be independent of the character of roll surface when "smooth" rolls are used, but dependent on degree of pressure employed.

The effect in practice may be well exemplified by the results of Table IV, which show the compositions and character of various grinds of a given semolina (*A*-roll feed) on smooth rolls. The untreated

TABLE IV

THE PERCENTAGE AMOUNTS AND MALTOSE FIGURES (SHOWN IN BRACKETS) OF THE SIEVE-SEPARATED FRACTIONS FROM GENTLE, MEDIUM, AND HEAVY GRINDS, RESPECTIVELY, OF SEMOLINA (*A*-REDUCTION) WITH SMOOTH ROLLS

The Maltose Figures of Corresponding Fractions under Different Grinding Pressures Reflect the Operation of the Internal Factor

	Percent of sample over					Flour (through 10 s.)
	24 g.g.	32 g.g.	1 s.	5 s.	10 s.	
Corresponding size of sieve aperture (*mm.*)	0.860	0.605	0.390	0.270	0.135	—
Feed ("primary" semo.)	—	16.0	73.0	8.8	1.0	1.0
Test No. 1 Gentle grind (low release with smooth rolls)	—	2.3	15.2 (0.78)	24.5 (0.57)	42.0 (0.53)	15.8 (0.98)
Test No. 2 Rather heavier	—	1.2	8.4 (1.19)	14.8 (0.81)	51.1 (0.64)	24.7 (0.93)
Test No. 3 Very heavy	1.7	4.5 (3.3)	8.8 (2.14)	11.8 (1.36)	33.1 (0.89)	39.6 (1.02)

semolina had maltose figure 0.37%. The maltose figures of the fractions of the grinds are shown in brackets beneath the corresponding figures for granulation as shown by the sieving analysis.

It will be seen that from this given sample of primary semolina (of maltose figure 0.37%) a series of products with maltose figures ranging from 0.53% to 3.3% may be produced through one rolling with smooth rolls. The immediate point is that the maltose figures of the coarser fractions of the grinds ascend rapidly as the pressure (and flour release) is increased, and the coarser they are the more they are affected in this way. For example, with the fraction through 32 g.g. over 1s., the "internal factor" has operated markedly in the case of the very heavy grind of Test No. 3, comparatively little in the light grind of Test No. 1. Put in another way we may enunciate the principle that when the reduction roll pressure is light the proportion of the total starch damage made which is borne by the coarser particles is low; as the roll pressure is increased the proportion borne by the coarse particles increases rapidly.

The operation of the internal factor in the way described may be confirmed microscopically.

The Internal Factor: Microscopical Demonstration

Generally speaking the examination of, and formation of conclusions regarding, large particles of endosperm is not straightforward because

the thickness of the mass prevents the penetration into the interior of stain (which is seized by the outermost gluten), cuts down the light for photographic purposes, and of course the occurrence of granules etc. at different depths interferes with a clear view of those above and below. The difficulty can in effect be overcome through a process of gradually and progressively dispersing or disintegrating the particle, the process being carefully and ceaselessly watched as it proceeds under the microscope. The dispersal is effected through a gentle to-and-fro motion of the cover slip over the slide with an absolute minimum of pressure.

The following observations were made on coarser particles from a smooth-roll grind somewhat similar to those through 32 g.g. over 1 s. of Test No. 2 in Table IV. The maltose figure of the feed in this case was 0.56 and that of the coarse particles in question in the grind 1.50. For convenience we will speak of "feed particle" and "grind particle" as meaning particles of the same nominal size taken from the feed and grind respectively. The grind particles are all more or less flattened though to very different extents according to their size. If the moderately flattened, discrete particles which represent the major part of the endosperm present are viewed after a short soaking in the stain, without any mechanical disarrangement, the appearance is much as shown diagrammatically in the sketch:

Relatively thin margin, e. g. one granule thick
Particle relatively thick here
A large flaked colony of "ghosts"
Small colony of "ghosts"
A few individual "ghosts"
Medium sized colony

It is noteworthy that what external damaged starch there is occurs at the edges of such particles and not on the upper or lower flattened surfaces of the thicker central part.

The actual appearance of such a particle is shown in Plate 6A and that of the feed particle (of maltose figure 0.56) in Plate 6B. No very great difference is apparent in this state of the particles apart from the presence of peripheral "ghosts" in Plate 6A and the contouring, a particle of unrolled semolina being thick throughout without any thinner margin.

The case is very different, however, when the cover slip is gently worked so as to cause gradual disintegration of the particle. The

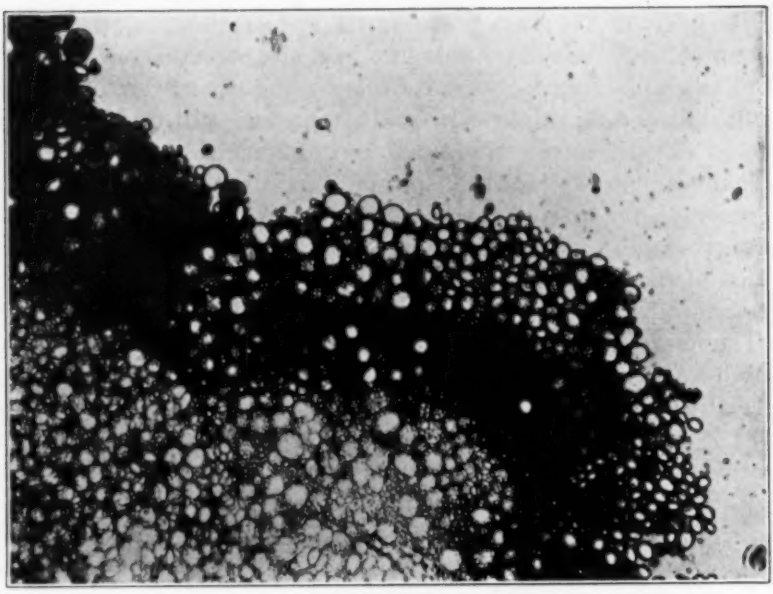


PLATE 6A

A grind particle (stained with congo red).

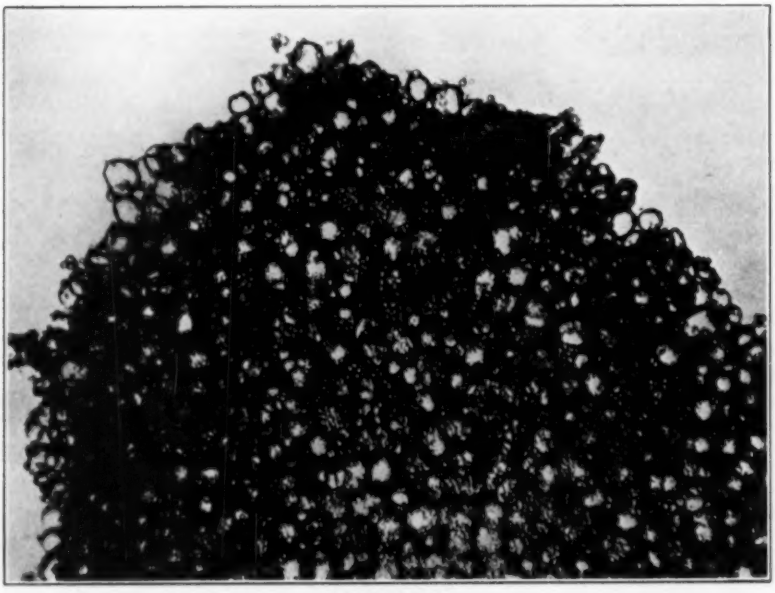


PLATE 6B

A feed particle (stained with congo red).

starch granules are gradually freed from the delicate gluten meshwork, teem towards the outside and are dispersed in the surrounding liquid. (Incidentally the "ghosts" behave somewhat differently in this respect from the sound granules as has been mentioned earlier. For instance, in working a particle a patch of "ghosts" hung together astonishingly well whilst the neighboring sound granules were washed freely away.) The gluten in turn consolidates itself forming very tangible rubbery rolls which are gradually freed from starch granules.

An idea of the true state of the starch in the interior of the grind and the feed particles, respectively, is given by Plates 7A and 7B. The large patches of apparently dark material in both photographs are lumps of deeply stained gluten. The starch in process of being dispersed in Plate 7B from the particle of original semolina is entirely sound. The process has been carried to an advanced stage in the case of the subject of Plate 7A; here the grind particle has been practically completely disintegrated, the lumps of gluten being relatively free from starch, and all the granules in the field were originally well in the interior of the particle. Many "ghosts" are to be seen amongst the sound starch granules. The important thing, however, is the constant watching of the process from the beginning of gentle dispersion onwards, the result leaving no doubt that, apart from the "ghosts" seen on the edges of the particle at the start, all those in the field of view *are from zones in the interior of the particle.* To make this clear it must be pointed out that when even strong stain solution is used in making up the slide, the stain is absorbed by the "ghosts" situated at the edges described and by the gluten of the outer layer all round the particle so that only water penetrates to the interior. On working the cover slip, the starch granules constantly streaming out from the interior of the particle include periodically batches of "ghosts" *which are unstained,* having been in contact with water only in the interior of the particle. Only when they reach the dye present in the liquid surrounding the particle do they at once take up the stain.

We are fortunate in being able to illustrate well this rather "tricky" matter through having secured a clear photo at a critical moment in the early stages of the dispersal of a fairly flattened grind particle. In Plate 8A there is a thin strand of gluten running in a diagonal direction which formed quickly and served admirably to protect the starch granules coming from the interior (situated towards the top and left-hand corner) from the stain in the outer liquid encroaching from the lower right-hand corner. To the left of the strand may be seen several "ghosts" as yet unstained, some of which are in a small colony which has moved out bodily amongst the freely scattering

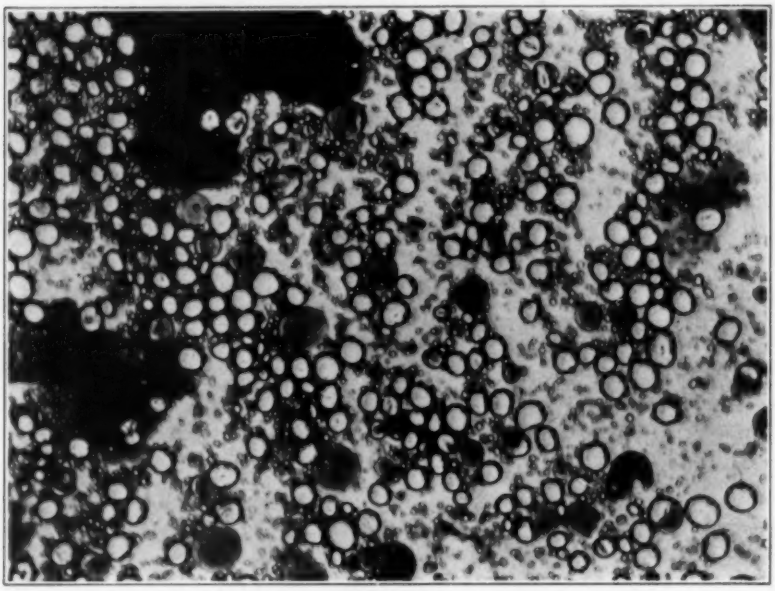


PLATE 7A

The grind particle after completion of "working" (stained with congo red).

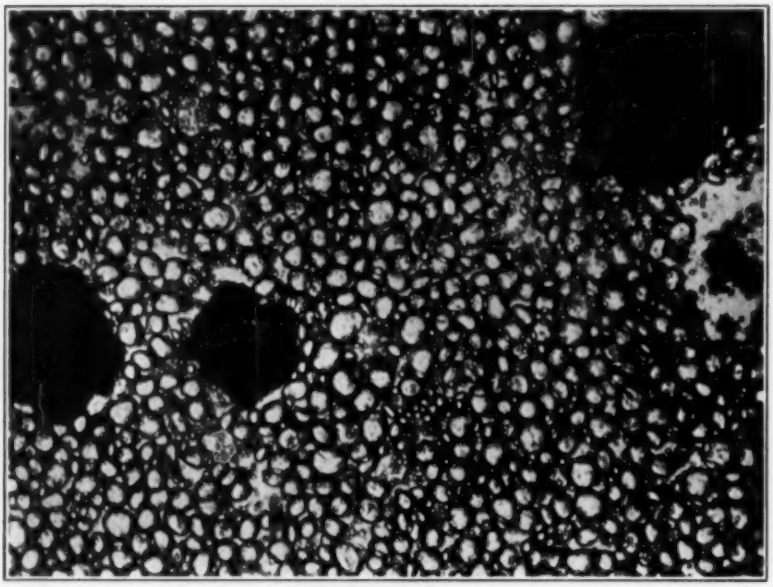


PLATE 7B

The feed particle after completion of "working" (stained with congo red).

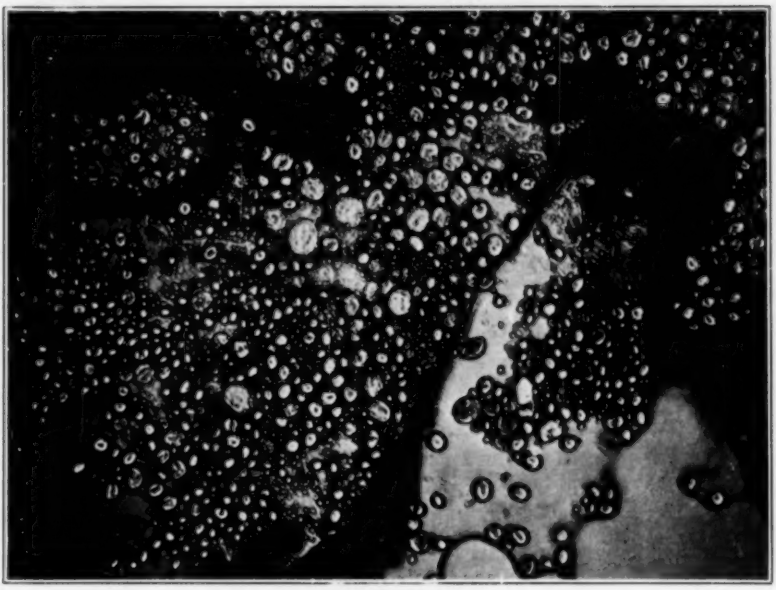


PLATE 8A

A grind particle after a little "working" (stained with congo red).

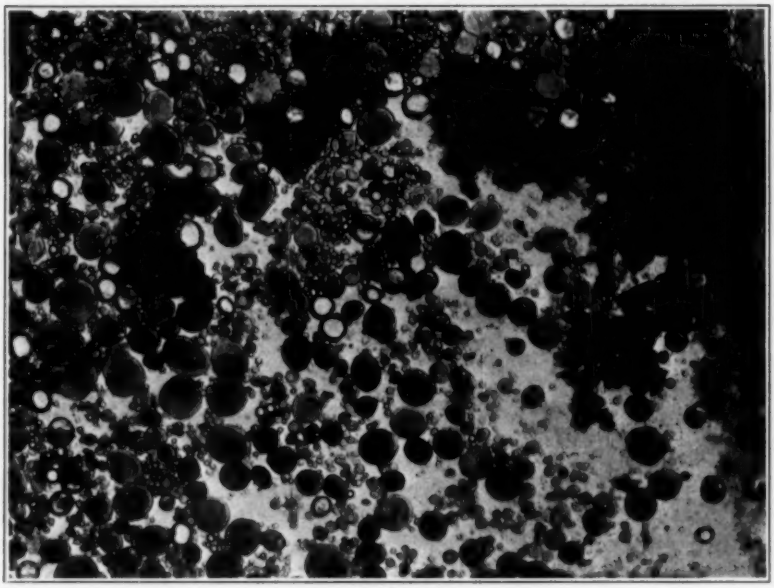


PLATE 8B

A particle of tail stock grind after "working" (stained with congo red).

sound granules. In marked contrast there is a number of stained "ghosts" on the right-hand side of the strand which initially were attached to the edge of the particle.

High Maltose Figures in Low-Grade Reduction Flours and "Tail Stocks"

If grind particles constitute a feed passing through smooth rolls a second time, any that escape disintegration will be very much flattened and will carry very severe internal damage. In this way the effect of the "internal factor" is cumulative, the maltose figure of such particles increasing markedly after each rolling.

With such particles there is no difficulty microscopically in obtaining evidence of the damage. They are really large flakes and are relatively thin so that the stain is able to penetrate fairly readily. On disintegrating a little the appearance is very striking as is shown by Plate 8B, which shows the case of coarse particles from a second consecutive grind with smooth rolls, the maltose figure being 4.3%. Very few of the larger granules are present except as "ghosts."

The practical consequence of this phenomenon is not difficult to understand. When coarse semolina is reduced in practice (on *A* reduction) the tails of the grind pass on to rolls further down the reduction system where they are again rolled. Now it is worth considering briefly the effect of further reduction on particles which have already been subjected to the "internal factor" as a result of having once been rolled. The maltose figure of the products from the second rolling (*i.e.*, the reduction of "tail stock") is a resultant of the following: (*a*) the breaking up to a greater or less extent of strained, damaged zones, *i.e.*, of colonies of "ghosts," from the interior of the large particles; (*b*) the action of the surface factor in which as before different types of roll surface behave differently and characteristically, but which is less than in the case of primary semolina owing to the mechanical weakening, already referred to, of the structure of the particle. That is to say, less resistance is offered to the scraping off of flour.

There is no need here to go into detail regarding the interplay of these factors under different conditions of reduction, but it may be stated that the effect usually is as follows: The flour formed is no lower than the feed in maltose figure and may be rather higher. The dunst is always appreciably higher in maltose figure than the flour, a fact which differentiates the operation from the reduction of primary semolina where the dunst tends to be appreciably lower in maltose figure than the flour (*A* roll). The difference is occasioned by the tendency of the "ghosts" liberated from the interior of the large particles to

adhere and remain together in comparatively large colonies. Relatively far more of the damaged starch from this source therefore remains in the dunst than is able to get through the flour silk.

Since in the reduction of "tail stock" the feed is high in maltose figure, the flour released is necessarily high also. This is the only explanation of the high maltose figures frequently found in very low-grade flours—contrary to the opinion sometimes expressed that such flours are high in maltose figure because heavily contaminated with offal particles or because they are very fine. Moreover, if the grinding pressure is increased on the head rolls of the reduction system the effect will at once be to raise the maltose figures of certain flours made further down the mill because under such conditions the maltose figure of the head-roll tail stock is greatly raised and therefore, also, the feed to the later reductions.

By way of partly summing up, it is possible to enunciate the following principles without which fluctuations of maltose figure, to be observed in the course of the milling process, are unintelligible:

1. Intermediate reduction stock carries damaged starch which becomes liberated during a subsequent release of flour.

2. The damaged starch can be carried in this way to varying degrees. Therefore, in the redution of dunst, for example, quite similar conditions of rolling may give rise to flour of appreciably different maltose figures, according to the previous history of the feed. Thus dunst from *A* reduction with smooth rolls and moderate pressures may commonly have a maltose figure of 0.7%. If on the other hand rougher rolls are used and also rather heavier pressures the figure will commonly become 1.0% or even rather higher. Since this stock is a major constituent of the feed to *C* roll there will commonly be a difference in the two *C* feeds of approximately 0.25% in maltose figure. The writer has found as a convenient rule in such cases that, other things being equal (identical operation of the *C* roll), the difference may be reckoned as added to the maltose figure of the flour made during the dunst reduction. If the conditions are such that in the first case the *C* flour has 1.4% maltose figure, then in the second case it will have 1.65% maltose figure and so on. These observations lead, together with the considerations of the "internal factor" already discussed, to the third principle.

3. The maltose figures of flours made at various points throughout the reduction system are influenced through changes made in the head reductions.

It is interesting now to consider the implications of the theory of the two factors in regard to the conditions of operation of the coarse

semolina reduction (*A* rolls), and to see how it fits with what is found in practice.

The Surface Factor

The surface factor—the scraping or attritional factor—should obviously be dependent for its extent on the roughness of the roll surfaces and on the differential speed with which they are driven. One would expect these to affect considerably the proportion of "ghosts" in the flour which consists mostly of the stuff torn off the surfaces of the large particles. One would expect them to affect the dunst liberated to a much smaller effect because the extent of surface produced is so much less in relation to mass than in the case of the flour. One would not expect them to affect appreciably the maltose figure of the coarser fractions of the grind because the starch damage here is chiefly in internal zones as a result of compression.

That these expectations fit in with practice will be seen from the results of comparative reduction tests on a sample of coarse semolina (*A*-roll feed) given in Table V. Maltose figures are shown in brackets beneath the corresponding figures for granulation according to the sifting analysis.

The very great effect of the matt rolls at the higher differential on the maltose figure of the *A* flour will particularly be noticed. Gen-

TABLE V

THE MALTOSE FIGURES (SHOWN IN BRACKETS BENEATH THE CORRESPONDING FIGURES FOR PERCENTAGE COMPOSITION AS SHOWN BY SIFTING ANALYSIS) OF THE FLOUR AND OTHER FRACTIONS OF THE GRIND OF SEMOLINA (*A*-REDUCTION) MADE WITH SMOOTH ROLLS, AND WITH VERY COARSELY MATT-SURFACED ROLLS AT DIFFERENT DIFFERENTIAL SPEEDS

	Percent of sample over					Flour (through 10 s.)
	24 g.g.	32 g.g.	1 s.	5 s.	10 s.	
Corresponding sizes of sieve apertures (*mm.*)..	0.86	0.605	0.39	0.27	0.135	—
Feed (untreated semolina).....	12.4	44.3	37.3	0.5	3.5	2.0
Grind with smooth rolls 1.27/1 differential.....	3.3	9.3 (2.12)	16.0 (1.49)	16.1 (1.00)	35.6 (0.85)	19.0 (1.40)
Grind with very coarse matt rolls at 1.27/1 differential.....	1.0	8.0 (1.72)	19.8 (1.21)	15.1 (0.90)	35.0 (0.89)	20.9 (2.34)
Grind with very coarse matt rolls at 2.1/1 differential.....	1.3	8.0 (1.74)	21.5 (1.12)	14.2 (0.95)	35.3 (0.98)	19.9 (3.93)

erally speaking the effect of differential is the more marked the rougher the roll surface and, as might be expected, it is progressively greater the greater the differential.

The very coarsest fractions of the grind tend to be lower in maltose figure with very rough rolls because a given release of flour can be made with rather less roll pressure owing to the greater attritional power of the surface. There is therefore less flattening of the coarsest parts of the grind and smaller production of internal damage.

Effects of Increase in Grinding Pressure in the Reduction of Coarse and Fine Particles

Returning now to consideration of the "internal factor" the data of Table IV have already shown sufficiently well how, as regards the coarser fractions of the grind, the use of increased roll pressure produces markedly increased starch damage in the expected way. What however is the effect on the maltose figure of the *A* flour of increasing the *A* roll pressure? It is strikingly characteristic of the reduction of coarse semolina particles that the maltose figure of the *A*-roll flour is substantially unchanged through variation in rolling pressure. This is shown well in Table IV. It is equally characteristic of this reduction that the *A*-roll flour is not made finer in granulation through increasing the rolling pressure; in fact it tends to be made grittier and larger in average particle size. This will be explained presently (see next section).

These considerations apply to the reduction of *coarse* endosperm particles. There are significant differences in the reduction of *finer* particles such as middlings or dunst, for example *C*-roll feed.

In the reduction of middlings the effect on starch damage of variation in differential roll speed is almost negligible compared with that in the reduction of coarse particles but the effect of the nature of the roll surface *per se* is still appreciable though somewhat diminished. It is in the effect of roll pressure that the reduction of fine stock is most sharply differentiated from that of coarse.

Table VI shows the effect on maltose figures (shown in brackets) of the fractions of the grinds produced by rolling middlings with smooth rolls (at 1.27/1 differential) set to various pressures. The maltose figure of the feed as a whole was 0.61%.

It should be explained that a thorough mechanical sifting test is used to analyze the samples and that the release of flour described as high in the table (test No. 74) is actually no more than is often found in commercial practice with *C* reduction. The small percentage found over 1 s. under these conditions represents flake production which always occurs to some extent when smooth rolls operate on fine stocks

TABLE VI

PERCENTAGE AMOUNTS AND MALTOSE FIGURES (IN BRACKETS) OF THE SIEVE-SEPARATED FRACTIONS FROM GENTLE, MEDIUM, FAIRLY HEAVY AND HEAVY GRINDS, RESPECTIVELY, OF MIDDLINGS (*C*-REDUCTION) WITH SMOOTH ROLLS The Trend of the Figures Is to Be Compared with That of Table IV (Dealing with Semolina)

		Percent of sample over			Flour (through 10 s.)
		1 s.	5 s.	10 s.	
Corresponding sizes of sieve apertures (*mm.*)		0.390	0.270	0.135	—
Test No.	Conditions of rolling				
—	Feed	—	11.0	76.0	13.0
71	Low release	—	4.4 (1.33)	71.0 (0.67)	24.9 (0.82)
70	Medium release	—	3.4	46.0 (0.85)	50.3 (0.96)
72	Fairly high release	1.0	3.1	29.1 (1.24)	66.7 (1.12)
74	High release	1.0	5.8	23.9 (1.78)	70.0 (1.26)

at high pressures. In fact under such conditions the dunst particles in the grind are flat and they and many of the flour particles are simply flakelets of varying size.

As seen from Table VI, in the reduction of fine particles at low releases (gentle pressures) with smooth rolls there is very little damage done to the starch. The increment in maltose figure (difference between that of the feed and that of the particular fraction of the grind) is small in the case of the flour. This is a marked differentiation from the behavior of coarse particles where, even with the gentlest reduction using smooth rolls, the increment in the case of flour is large (say 0.7) and in fact as high as under conditions of severe reduction.

Further, with fine particles, the maltose figure of the flour rises progressively as the pressure and release are increased. The rise becomes more rapid as the release increases.

The maltose figure of the remainder of the grind, chiefly the dunst, also rises even more rapidly. The "increment" in the case of the dunst is practically non-existent at low releases but it greatly exceeds that for the flour at high releases.

How are these differences between the reductions of coarse and fine particles to be explained? It should be noted that reduction of middlings differs markedly from that of coarse semolina in that increasing rolling pressure affects the granulation of the flour. The flour which is coarsely granular at lower releases becomes progressively and markedly finer as the release is increased.

The radical difference between the effect of varying pressure on the maltose figure of the flour made during a rolling of coarse stock on the one hand, and of fine stock on the other, must not, however, be attributed simply to a consequence of the constant granularity in the one case and the increasing fineness of the flour in the other. It is due, rather, to the different underlying factors which in turn give rise to the different effects on granularity.

Interpretation of the Different Effects Observed in Reduction of Coarse and Fine Particles

It appears that the essential features of the reduction of milling stocks may be viewed in this way: a particle of coarse semolina may be, let us say to simplify matters, roughly cubical in shape and, say, 0.5 mm. long each side. On crushing under normal conditions as in *A* reduction it may break up principally into, say, 16 pieces, the ratio of whose length of side to that of the original particle will, of course, be the cube root of 1/16, namely, 1/2.5. That is to say, the new pieces will have a length of side on the average of 0.2 mm., which would correspond to stock overtailing a 10 silk but passing through a 60 g.g. Now if the pressure on the rolls is considerably increased, *i.e.* the grind made lower, the original particle will break up not into 16 but principally, say, into 32 fragments, so that the average size of the particle formed has moved further down. In addition to this sort of process, in *A* reduction there is always a certain amount of quite fine flour produced which must be regarded as scraped or torn off the surface of the large particle through contact with the roll surface, under the influence of the differential speed.

When the grind is lowered and the pressure increased in the case of *A* reduction the original flour is thus reinforced by a considerable amount of relatively coarse particles, which are dunst as it were moved or reduced a stage down. In other words, the increased pressure has caused many of the particles of the chief product, the dunst, to be smaller than they were in the first place: not very much finer, but fine enough now to pass through the flour silk. It is in this way that the, at first sight, surprising result is obtained that increasing the pressure on *A* roll, so that the release of *A* flour increases from, say, 18% to, say, 35% or even more, does not cause the *A* flour to be at all finer and softer but may even make it feel more gritty. What is the effect of the increased pressure on maltose figure of the *A* roll flour? It has been briefly mentioned already how the dunst from *A*-roll grind, the stuff of intermediate granularity, has the lowest maltose figure of all parts of the grind. Its particles largely escape the internal damage (production of "ghosts" in zones inside the particle) which happens

to the coarser overtails. They do not receive such severe peripheral damage as the fine flour produced through the action of the roll surface in tearing material away from the surface of the large particles. At the same time, any peripheral damage which they do receive has much less effect on the maltose figure, because, owing to the greater particle size compared with the flour, and lower ratio of surface area to total mass, the damage per unit weight is so much less. Much the same applies to the material now under discussion which, when the pressure on *A* rolls is increased, becomes in effect dunst which is just fine enough to pass through the flour silk. The amount of damaged starch it brings into the flour is small enough in relation to the total amount of sound starch introduced not to cause any increase in maltose figure.

It is possible too that when increased roll pressure is applied to *A*-roll feed, a certain amount of the finer material liberated originates directly from the interior of the large particles of endosperm constituting the bulk of the semolina, and as such has not been influenced at all by the surface action largely affecting the flour made under conditions of lighter rolling. Either of these factors ensures that the maltose figure of the *A*-roll flour does not increase upon lower grinding.

In *C* reduction one starts in a much lower octave, as it were. The feed as a whole is relatively very near in particle size to the mesh apertures of the flour silk, and when any given particle breaks up on high grinding into a certain limited number of smaller pieces the whole of these pass through, say, a 10 silk, the flour so formed being quite coarse and granular.

As the grind of *C* roll is lowered, and the release made greater, the bulk of the particles formed become progressively and relatively fairly uniformly smaller in size. The *C* flour becomes finer and softer to the feel. Not only so, but owing to the greater violence (greater energy expended) more and more of the starch granules situated around the surfaces of these particles become damaged (converted into "ghosts"). A good idea of the change occurring may be had from a study of Plate 9, which shows microphotographs (magnification approximately ×120 diameters) of two of the *C* flours of Table VI, in congo red stain. Photograph A shows a typical view of flour No. 71 corresponding to a release from the *C*-roll feed of only about 25%, with a maltose figure of 0.82%. Photograph B shows flour No. 74, corresponding to a 70% release, with a maltose figure of 1.26%. It will be seen how on the whole the flour particles are very considerably smaller in the latter flour. The important thing with respect to maltose figure is the presence of a greatly increased number of "ghosts" in flour No. 74.

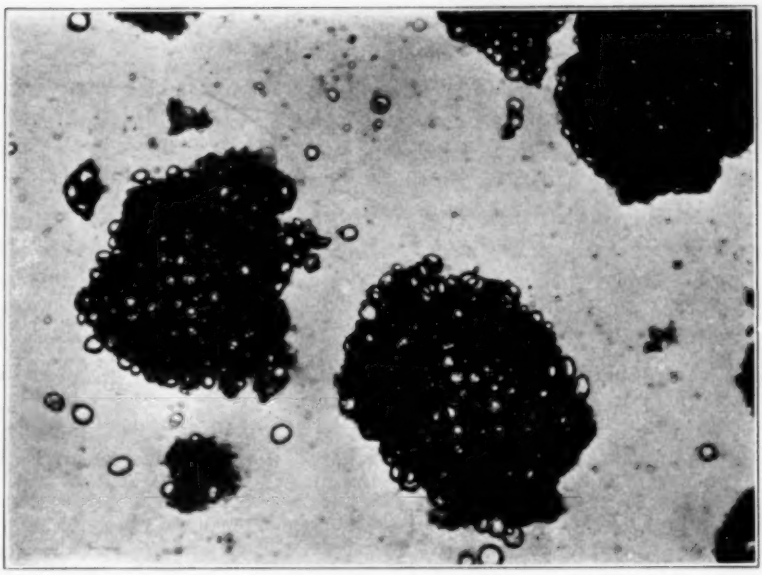


PLATE 9A

Particles of flour made by reducing middlings at different roll pressures. Release 25%, maltose figure 0.82.

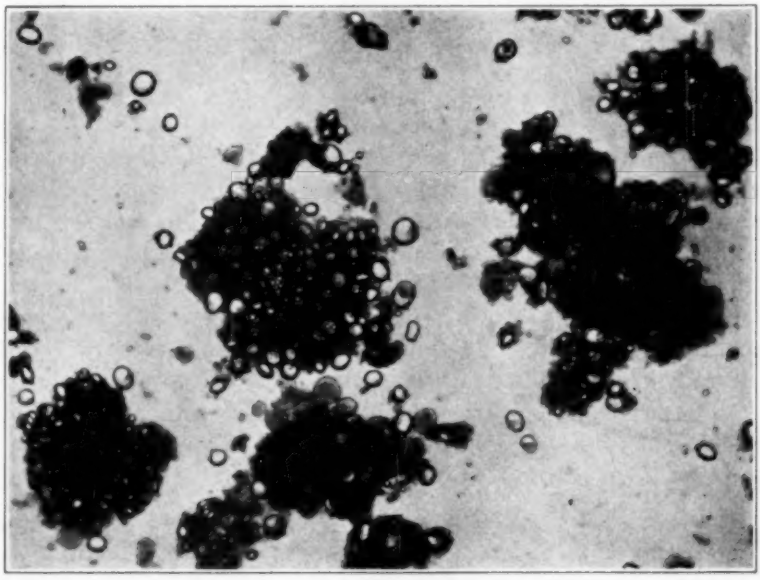


PLATE 9B

Release 70%, maltose figure 1.26.

Production of Damaged Starch as Affected by Type and Hardness of Wheat

This paper cannot well be concluded without a brief reference to a practical consequence of the considerations given. The production of "ghosts" in the interior of large particles during reduction has been shown to be due to the strain resulting from compression. Naturally the extent of this strain will depend on type and condition of wheat. To a considerable extent this is true also of the operation of the surface factor. Generally therefore other things being equal the maltose figure of the flour becomes higher the harder the wheat. This is markedly so under conditions of severe milling because the internal factor then plays a preponderating part. In illustration Table VII may be given.

TABLE VII

MALTOSE FIGURES (SHOWN IN BRACKETS BENEATH THE CORRESPONDING FIGURES FOR PERCENTAGE COMPOSITION AS SHOWN BY SIFTING ANALYSIS) OF THE FLOUR AND OTHER FRACTIONS OF GRINDS, UNDER COMPARABLE CONDITIONS, OF SEMOLINAS FROM HARD AND FROM SOFT WHEATS

	Percent of grind over					Flour (through 10 s.)
	24 g.g.	32 g.g.	1 s.	5 s.	10 s.	
Corresponding lengths of sieve apertures (*mm.*)	0.860	0.605	0.390	0.270	0.135	—
Grind of Manitoba semolina	1.0	6.0 (3.67)	12.4 (2.68)	18.8 (1.74)	40.4 (1.10)	20.4 (1.53)
Grind of soft-wheat semolina	1.3	4.8 (1.76)	12.6 (1.19)	17.5 (0.81)	43.6 (0.64)	20.2 (0.93)

Table VII shows the composition according to sifting analysis and (in parentheses) the maltose figures of the respective fractions of the grind in the cases of (1) a reduction of coarse semolina from all Manitoba wheat, and (2) a similar reduction of coarse semolina from soft wheat (ordinary Yeoman). The feeds (untreated semolinas) were of similar granulation.

It will be seen that, as a result of the one grind, the Manitoba semolina gave a series of products ranging from 1.10% to 3.67% in maltose figure, and the soft wheat from 0.64% to 1.76%. The range in the one case is more than double that in the other, as a result of the different degrees to which the "internal factor" operated in the two cases.

Summary

All flours contain a certain proportion of their starch granules mechanically damaged as a result of milling processes. This is a fortuitous advantage, for in the absence of such damaged granules no

appreciable diastatic action would occur in the suspensions or doughs made from flour from ordinary sound wheats. Wheat-starch granules are subject to more than one type of mechanical damage but it is shown that only one type results from roller milling processes. The nature of this particular type of damage is discussed. Owing to their characteristic microscopical appearance in the unstained state, granules affected in this way have been dubbed "ghosts"—a not inconvenient way of avoiding a plurality of words in referring to them.

It is shown that the enzymes responsible for diastasis in sound wheat products must be regarded as uniformly distributed throughout the endosperm. Diastasis in a water suspension is not affected by particle size *per se*, and in flours and intermediate stocks variously milled from a given wheat the maltose figure is a measure of the number of "ghosts" present.

"Ghosts" are present in flours to very different extents according to the conditions of milling. The conditions under which they are produced during milling are analyzed. Two broad factors operate: the "surface factor" and the "internal factor": the one dependent on the shearing or scraping of material from particle surfaces, the other on the crushing or partial flattening of larger particles. It is characteristic of the type of damage to the granule in "ghosts" that it is produced by both the shearing and the rapid crushing action encountered in roller milling. The distinction between the two factors however is important in determining the "ghost" content (or maltose figure) of the final straight-run flour.

With primary stocks of different particle sizes (primary means coming directly from the breaks, *i.e.*, unreduced), the maltose figure remains low as the particles become finer until the flour itself (say throughs of No. 10 silk) is reached. It is shown that the operation of the "internal factor" causes the coarser fractions of a reduction roll grind to be relatively high in maltose figure, because their particles carry *internal* zones of damaged starch. These result from the strain set up in the particles as a result of the partial flattening during the rolling. When coarse primary stock (such as *A*-roll feed, in British milling terminology) has passed once through smooth rolls, the maltose figures of the coarser fractions of the grind are thus relatively high, whilst those of the intermediate fractions (dunst or *C*-roll feed) are low. The heavier the roll pressure and the coarser the particles concerned, the higher the resulting maltose figure, owing to the greater intensity of operation of the internal factor. The flour produced from such coarse particles (*A*-roll flour) has a more or less high maltose figure which however is not affected through change in grinding pressure.

Its content of "ghosts" is due to the surface factor only, and its maltose figure accordingly is markedly affected through nature of roll surface and differential roll speed, factors without influence on the internal factor, which causes the damage in the coarser fractions of the grind.

In general the important principle appears that any incompletely reduced particles (intermediate reduction stock: dunst or tails) carry damaged starch which becomes liberated, in addition to that freshly made, during a subsequent release of flour, *i.e.* when the intermediate stock in question becomes the feed to a later reduction. It is shown that this results in, and is the sole cause of, relatively very high maltose figures of flour and other stocks from later reductions, and that the magnitude of such maltose figures depends sensitively on the conditions of the earlier, or head, reductions. In general the effect of the internal factor is cumulative, the maltose figure of the coarser particles which may escape disintegration increasing markedly after each rolling.

It is explained that there are significant differences with respect to maltose figure between the reduction of finer particles (middlings or dunst, *e.g.* *C*-roll feed) and that of coarse (semolina, *e.g.* *A*-roll feed). In the reduction of middlings the effect on starch damage of variation in differential roll speed is almost negligible compared with that in the reduction of coarse particles, but the effect of the nature of the roll surface *per se* is still appreciable though somewhat diminished. It is in the effect of roll pressure that the reduction of fine stock is most sharply differentiated from that of coarse. With gentle reduction of fine stock few "ghosts" are produced, but increase in the roll pressure leads to a rapid rise in maltose figure of flour and a still more rapid rise in that of the remainder of the grind, chiefly the dunst. An explanation is advanced of the mechanism underlying the essential differences between the reduction of fine and coarse particles.

An interesting practical consequence of the considerations given is in the case of different types of wheat. The extent of operation of the internal factor depends on type and condition of wheat; to a considerable extent this is true also of the surface factor. Generally therefore the maltose figure of the flour becomes higher the harder the wheat. This is markedly so under conditions of severe milling because the internal factor then plays a preponderating part. Differences in diastatic activity between flours from different types of wheat are thus not *necessarily* due to differences in amylase content or in starch "susceptibility." They are at least partly to be attributed to differences in the physical hardness of the endosperm as affecting the extent of the damage to the starch during milling.

Acknowledgment

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A BIOCHEMICAL AND TECHNOLOGICAL STUDY OF PUNJAB WHEAT VARIETIES[1]

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Wheat is the mainstay of Punjab agriculture. The importance of Punjab as a wheat-growing country can well be imagined by the fact that of all the provinces of India, taken singly, it has the largest acreage under wheat. It not only produces enough wheat for home consumption, but is the premier wheat-exporting province. Of the total quantity of wheat exported from all India, Punjab contributes the major portion. The merits and shortcomings of Indian export wheat, whatever they may be, are largely those of the Punjab wheat.

Area, Production, Yield

During the decennial period ending with 1934–35, out of an average of 33.2 million acres on which wheat was grown annually in India (including native states), 9.4 million acres or 28% belonged to the Pun-

[1] Condensed from a thesis presented by Rattan Singh to the Graduate School of the University of Minnesota in partial fulfillment of the requirements for the degree of Master of Science.

Fig. 1. Outline map of India.

TABLE I

ACREAGE AND PRODUCTION OF WHEAT IN INDIA AS COMPARED TO THAT OF THE PUNJAB, INCLUDING NATIVE STATES IN BOTH CASES

	Acreage—millions of acres		Production in millions of tons	
Year	India	Punjab	India	Punjab
1925–26	31.0	9.5	8.8	2.9
1926–27	31.8	9.4	9.1	2.9
1927–28	32.7	9.0	7.9	2.3
1928–29	32.5	10.0	8.7	3.1
1929–30	32.2	10.0	10.7	3.8
1930–31	32.7	9.3	9.5	3.1
1931–32	34.3	9.1	9.2	2.8
1932–33	33.5	8.6	9.6	2.8
1933–34	36.6	9.8	9.6	2.8
1934–35	35.0	9.0	9.9	3.0
Average	33.2	9.4	9.3	2.9
Percent of total	—	28.3	—	31.1

jab. Of these 9.4 million acres, 5.2 million or 55.3% is irrigated and 4.2 million or 44.7% is *barani* or rain-dependent.

The wheat area of the Punjab ranges annually between 9 and 10 million acres. This variation occurs in the *barani* area which, of

necessity, has to depend upon the rains. The irrigated area, however, remains almost constant from year to year (Table II).

TABLE II

EXTENT OF IRRIGATED AND UNIRRIGATED WHEAT ACREAGE IN THE PUNJAB

Year	Wheat acreage in the Punjab, in millions of acres	
	Irrigated	Unirrigated
1925–26	5.1	4.4
1926–27	5.0	4.4
1927–28	5.1	3.9
1928–29	5.3	4.7
1929–30	5.6	4.4
1930–31	5.4	3.9
1931–32	4.9	4.2
1932–33	5.0	3.6
1933–34	5.0	4.8
1934–35	5.2	3.8
Average	5.2 (55.3%)	4.2 (47.7%)

During the same period the average annual production was 2.9 million tons, which was almost 39% of the total production of 7.5 million tons for British India. If Indian states are included, then it becomes 31% of a grand total of 9.3 million tons. A diagrammatic representation of acreage and production of wheat in the different provinces of India is shown in Figure 2.

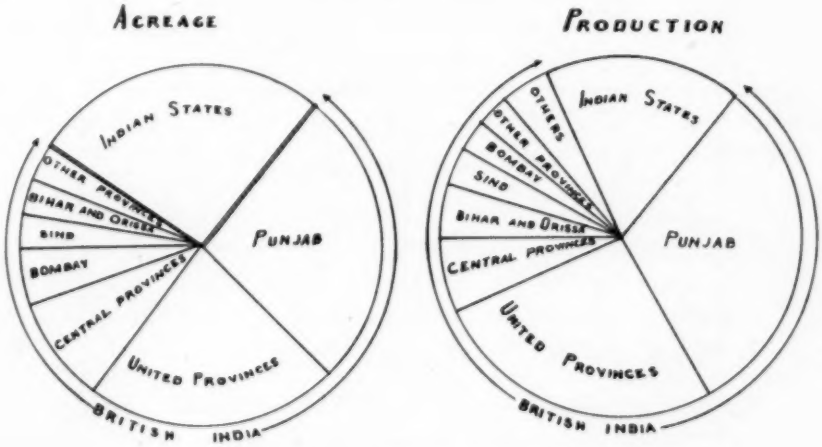

Fig. 2. Distribution of wheat production in India.

The average yield per acre of wheat in the *barani* area is estimated to range between $9\frac{1}{4}$ and $10\frac{2}{3}$ bushels (bushel = 60 lbs.). On the irrigated lands the yield is between 15 to 17 bushels, and the combined

average is between 12 and 14¼ bushels, *i.e.*, of the order of the United States. Though the average yield of the province as a whole is low, yields of 28 to 30 bushels from large-size blocks of a thousand acres or more of land in the new canal colonies are on record.

Internal Consumption and Trade

It has been estimated that something like 72% to 84% of the total production is retained in the province and largely consumed in the form of *chapaties* (unleavened pan-baked cakes). There are about a score of modern roller mills working in the province. A certain amount of fermented bread in the form of loaves is produced by the *bazar* bakers and other bakeries in the cities and is coming into use. The remaining 16% to 28% is exported either to other provinces, or out of India, chiefly through the port of Karachi (Figure 1) under the label of choice white Karachi or red Karachi wheat.

Figure 3 gives the per-capita consumption of wheat in the different provinces of India. This shows that in general the Northern parts of

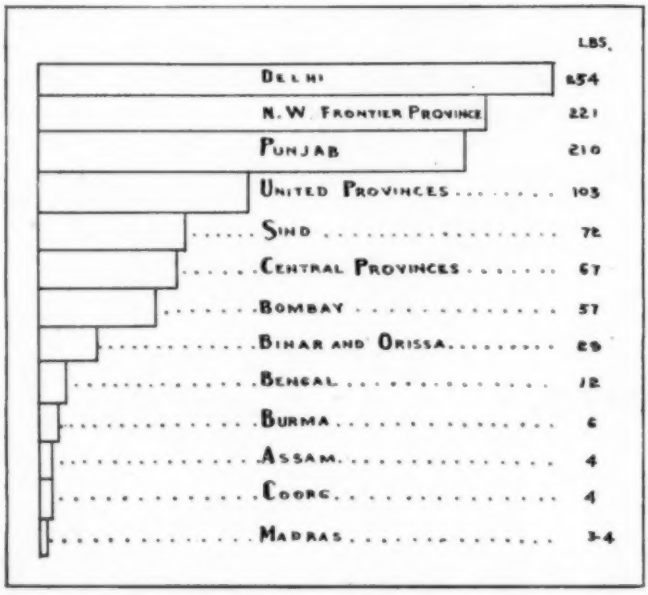

Fig. 3. Per-capita consumption of wheat in the provinces of India.

India produce more wheat and the consumption per capita is proportionately greater. This, however, declines towards the south or east where rice is the staple cereal crop. The lowest consumption per capita of wheat is in Madras.

Area and Location of the Punjab

The Punjab comprises an area of 133,000 square miles, of which 36,500 square miles belongs to Indian states, and is one of the provinces of India. Mexico, southern Arizona, Texas, Mississippi, Georgia, etc., are situated in about the same latitude. Gibraltar is only two degrees further north of the equator than the northern districts of the Punjab.

Lyallpur, one of the 30 districts, is situated in the canal colony area of the Punjab. It was founded about 45 to 50 years ago when this portion of the Punjab, till then an uncultivated waste, was brought under irrigated cultivation by means of "perennial" canals. It is the headquarters of the Department of Agriculture of the Punjab as well as the seat of the Punjab Agricultural College and Research Institute.

Climate and Soil of the Punjab

The Punjab is an inland province. Since it has a light soil and much of it is 500 to 600 miles from the sea, it is subjected to extreme variations of temperature. In June maximum shade temperatures have been recorded as high as 122.6°F. The recorded temperatures have been as low as 8° below the freezing point. This low temperature, however, does not do much harm because the cool mornings are usually followed by sunny days. Hot weather generally begins in February and becomes rather unpleasant in April when hot winds come from the west.

There are two well defined rainy seasons, one of which is known as the summer or monsoon and the other the winter rainy season. Monsoon season in Punjab begins about the end of June or the middle of July and ceases about the middle of September or earlier. The winter rains, from December to March, are more uniformly distributed. It is on the volume and distribution of both of these rainy seasons that the annual acreage and yield of wheat in the Punjab depend.

Of the 30 districts there are about a dozen which, being either mountainous or submountainous, have enough annual precipitation for ordinary agricultural purposes, *i.e.*, 25 inches or more. Elsewhere it is less than 25 inches and diminishes in some southwestern districts to as little as 5 to 7 inches. Deficiency of rainfall in these districts has been supplemented by irrigation from the perennial canals, however. To judge the extent of the area watered by canals consider that in 1868–69 about 1.37 million acres were irrigated while in 1918–19 and 1929–30 (including Indian States) about 9 and 12½ million acres were irrigated, respectively.

Rotation, Cultivation, Sowing, and Harvesting

Wheat in Punjab is almost always sown on land that has been left fallow for several months after a previous crop of cotton, sugar cane,

wheat, gram (*Cicer arietinum*), or a millet. In the hills, however, owing to the smallness of holdings it is sown after maize (*Zea mays*) and rice, without any fallow. Generally eight to ten plowings on land dependent upon rains and five to six on irrigated lands are given. Only a small portion of wheat land is manured directly and then with ordinary farmyard manure alone.

Sowing begins late in September or early in October and continues until the end of December or even January, by far the greater portion of the area being sown between October 15 and November 15. Though sown in the fall, Punjab wheats can be spoken of as spring wheats, since they are rarely subjected to severe winter frosts. Generally four irrigations of a total amount of 10 to 12 inches are given canal-irrigated tracts, including the one given for sowing of the crop. When wells are the source of irrigation water the crop receives two or three irrigations more since the individual applications are lighter.

Harvesting begins at the end of March and the threshing is finished by the middle of June before the monsoon breaks.

Varieties

Three species of wheats are grown in the Punjab: *Triticum sphaerococum*, *T. durum*, and *T. vulgare*. Of the first group seven unit-species belonging to seven different varieties have been separated. These were found in dry districts of the southwest and are no longer cultivated. Four unit-species of the durum group belonging to four different botanical varieties have been separated. Only one of these, *viz.*, type No. 1, belonging to the Malanopus variety, is economically important. Since the durums need plenty of moisture and rich soil, their cultivation is restricted to only two or three districts of the Punjab where there is enough moisture. Durum wheat is used chiefly in the manufacture of semolina, which itself is used in making *halwa*, a kind of pudding. Durum is not exported. The third group, *T. vulgare*, is by far the most important, for it occupies most of the wheat acreage of the Punjab. About 65 unit species belonging to 20 different botanical varieties have been separated from the local mixtures. Of the types isolated, about two-thirds are beardless and one-third bearded, but as a class the former does not yield as much as the latter.

Improvement of Wheat in the Punjab and the Purpose of this Investigation

In 1905 a beginning was made towards organizing an agricultural department on modern lines in the Punjab. At first 25 types of wheat were separated from the local wheat mixtures. Later surveys showed that local wheat crops were everywhere a mixture of types, of which

the most common were Types 11 (var. *erythroleucon*), 13 (var. *ferrugineum*), 14 and 15 (*erythrospermum*), 16 (*graecum*), 17 (*Delfi*), 24 and 25 (*albidum*).

Economic improvement of wheat by the Department of Agriculture was started by distributing wheat type No. 9 (var. *pseudo-Hostianum*). It gave good yields on the Department's farms, but did not do well on farmers' land and was replaced by type No. 11, referred to above. This type was decidedly better than the local mixture and on the average gave 1½ to 2 *maunds* (1 *maund* = 82⅔ lbs.) of grain more per acre. Official distribution of seed of type No. 11 started in 1913 and in 1924–25 it had reached 930,000 acres. Thereafter its cultivation has been declining in favor of another type called 8A belonging to var. *Turicucum*, which was first discovered as a single plant in a sheaf in one of the districts of the Punjab. This is a hard, amber-grained bearded wheat with reddish velvety chaff. Comparative varietal tests extending over a number of years conclusively showed that 8A yields 1½ to 2 *maunds* of grain per acre more than type No. 11 and more straw, which is a valuable fodder for cattle feeding in the Punjab. It was officially distributed to the farmers for sowing in 1919, and by the end of June, 1936, the area seeded to it was 3,161,700 acres. With the exception of Marquis, perhaps no other single product of wheat breeding anywhere has had so great a success as 8A. Numerous introductions of other wheat varieties have been made from Europe, America, Australia, Canada, etc. but not a single one has ultimately succeeded.

Although 8A still continues to be the premier wheat of the province, later observations showed that it had a few drawbacks:

(1) In the Punjab, maize and sugar cane are always sown on land which has previously been heavily manured with farmyard manure. When wheat follows maize or sugar cane it generally lodges. This was the case with 8A. Land of this level of productivity is not very extensive, however.

(2) Sometimes the farmer is compelled to seed his wheat as late as December or even January. This may be occasioned either by late rains in the *barani* area or by irrigation of the cotton crop, which is in the fruiting stage at that time, in preference to watering land for preparation of the seedbed for wheat sowing. Under such late-sown conditions it is desirable that the wheat be able to ripen in time; otherwise it is likely to be caught by the hot winds in early April, when the grain is still in the dough or hardening stage. This results in pinching of the grain and consequent diminution in yield. Being midseason in maturity, *i.e.* neither early nor late under the Punjab conditions, 8A takes full advantage of the normal Punjab wheat season.

Experience has shown, however, that this wheat when planted late is exposed to injury by hot winds and sometimes actually suffers when the hot winds set in earlier.

With a view to effecting improvement in some of the defects in 8A mentioned above, later researches have yielded three new wheats. These may be called the special-purpose wheats. These are C518, C591, and 9D. The first two are crossbred varieties and the last-named is a pure-line selection and belongs to the variety *pseudo-Hostianum*.

C518.—This is a fully bearded but comparatively short-awned amber-grained wheat with white, velvety chaff and grayish black awns. Ears are dense and erect. The straw is comparatively short and very stiff. It resists lodging to a very great extent, exhibiting this fault only on very rich soils. It yields better than 8A on lands of higher level of production, the combined average increase of 80 comparisons in six years 1929–30 to 1934–35 being $3\frac{3}{8}$ maunds of grain per acre more than 8A. When good land and an adequate supply of water are available, C518 gives very good yields. The highest yield per acre with an authentic record to date was 65 bushels (Singh, 1933). This was a calculated acre yield, however, from a $\frac{1}{72}$ acre plot.

C591.—This is fully bearded, amber-grained wheat with white, velvety chaff and grayish awns. This also resists lodging, though not as much as C518 but better than 8A. On good lands it yields better than 8A.

9D.—This wheat is a fully bearded wheat with white awns, white velvety chaff, and amber plump grains. It is recommended for sowing under the late-sown and *barani* conditions.

Chapati-making properties of these wheats.—As mentioned elsewhere most of the wheat produced in the Punjab is consumed in the form of chapaties (unleavened pan-baked cakes) prepared from whole-wheat meal. Therefore, in addition to high-yielding capacity and good milling and baking properties according to standards of European and other western countries, it is necessary, in order that it may meet with the approval of the farmers and general body of the consumers, that a new variety for distribution should possess satisfactory chapati-making qualities. Numerous tests in this direction have shown that all these wheats have good chapati-making properties. C591 is a superb wheat in this direction and gives perhaps the most tasty and excellent product. Second in the list comes 8A though C518 and 9D are more or less like it.

Area under improved wheats in the Punjab.—The total area sown to the improved wheats according to R. D. Singh [2] (personal communica-

[2] Cerealist, Punjab Agricultural College, Lyallpur, India.

tion to the author) up to 1936 has been estimated to be 4,661,700 acres, including, of course, 3,161,700 acres under 8A alone. This is about 50% of the total area of 9 to 10 million acres commonly sown to wheat in the Punjab.

Progress of wheat hybridizing work.—In order to evolve still better wheat having all-round good qualities, an extensive program of cross-breeding work is under way at the Botanical Farm, Lyallpur. During the six years beginning from 1929–30, 57 new crosses were made. These yielded about a hundred thousand (98,510) plants in the F_2 generation. Each of these plants was studied singly and the selection of the desirable ones made from year to year. Twenty-three stable wheat-type samples out of 31 shown in Table III are selections made from some of the above cross-bred material. These are selected on the basis of their disease resistance and high-yielding capacities. No final decision can be made, however, so long as their quality is not examined. The purpose of the present investigations, therefore, is to test their milling, baking, and other physico-chemical qualities according to the fermented-bread-making methods. These can then be correlated with their chapati-making values and high-yielding powers. Any one of these proving satisfactory in all these respects can be finally selected for releasing to farmers for sowing.

Material

A list of wheat types dealt with in the investigation is given in Table III. These wheats were raised on the Botanical Farm of the Punjab Agricultural Research Institute, Lyallpur, in 1935–36. Most of them were grown under irrigation. Some, however, were sown under *barani* conditions to test their suitability for such environment, because, as earlier pointed out, about 45% of the total wheat area in the Punjab every year is under *barani*. The irrigated plots were given four waterings and the *barani* only one and that merely to moisten the land for preparation of the seedbed for sowing and none thereafter. Rainfall during the year 1935–36, *i.e.*, from the first of July, 1935, to June, 30, 1936, was only 12.46 inches, which is 1.09 inches below the average of the 1926–35 decennial period. Of this precipitation, 5.33 inches occurred in the fallow period (monsoon season) and 2.6 in the growing period (winter rain), which was rather dry and moderately cold.

These types can be discussed under two different headings: (1) Serial numbers 1 to 8 are the duplicate samples of the four types, which are on the approved list of the Department of Agriculture and extensively grown by the farmers. (2) Numbers 9 to 31, with dupli-

TABLE III

DESCRIPTION OF THE WHEAT SAMPLES USED IN THESE TESTS

Serial No.	Type No.	Parentage	Conditions of growth	Remarks
1	8A	Varieties	Irrigated	Nos. 1–8 duplicated samples of types that are on the approved list of the Department of Agriculture, Punjab, and are being grown by farmers.
2	8A	on the	"	
3	9D	approved	*Barani*	
4	9D	list of the	"	
5	C518	Department	Irrigated	
6	C518		"	
7	C591		"	
8	C591		"	
9	C205	H.F.×9C	"	Nos. 9 to 31 are new cross-bred wheats under preliminary trials. Some of them are duplicate samples.
10	C206	"	"	
11	C207	"	"	
12	C207	"	*Barani*	
13	C208	"	Irrigated	H.F. = Hard Federation. In writing parentage, the female parent is shown towards the right and the male parent towards the left.
14	C209	"	"	
15	C210	C518×H.F.	"	
16	C212	C518×H.F.	"	
17	C215	C516×C591	*Barani*	C = Cross, meaning a hybrid.
18	C201	H.F.×9D	Irrigated	Irrigated means that the crop was irrigated by artificial means of irrigation, canal irrigation in this case.
19	C201	"	*Barani*	
20	C202	"	Irrigated	
21	C202	"	*Barani*	
22	C203	"	Irrigated	
23	C204	"	"	*Barani* means dependent on rains. In Lyallpur, where these wheats were raised, the land was irrigated only once for seedbed preparation and no irrigation was given thereafter.
24	C224	"	*Barani*	
25	C225	"	"	
26	C227	"	"	
27	C227	"	"	
28	C228	"	"	
29	C228	"	"	
30	C229	"	Irrigated	
31	C229	"	*Barani*	

cate samples in some cases, are all recent hybrids, selected from much cross-bred material. These are still in the preliminary stage of trials.

Numbers 9 to 14 have Hard Federation and 9C as their parents. The former is a selection obtained from Australia, said to lodge less and to possess good milling and baking qualities. 9C, however, is an indigenous selection. This too possesses good milling and baking qualities. Nos. 15 and 16 are selected from the progeny of C518 and Hard Federation. No. 17, *i.e.* C215, is a composite cross, having C516 as one of its parents. C516 wheat, while establishing its unquestionable superiority to all other wheats against which it was tested under conditions of higher level of productivity, happened to be of very poor baking quality and had to be discarded. Efforts have been made to improve its quality, however, by combination with a better quality wheat, *i.e.* C591. Nos. 18 to 31 are all selections from the progeny of a cross between Hard Federation and 9D. The latter does not possess

acceptable milling qualities, probably because it has a low gluten content. Moreover, it has poor standing power. With a view to improving on these poor qualities, it has been crossed with Hard Federation.

Table VI gives the 1000-kernel weights of the samples. Each figure is an average value of two determinations; the average of the 21 samples comes to 41.4 g., with a range from 33.8 to 48.1. This shows the heaviness and plumpness of the kernels and less variation in the kernel weight.

This exceeds the average value for all India wheats (Table V) by

TABLE IV

KERNEL WEIGHTS OF WHEATS GROWN IN MINNESOTA, 1911–1913 [1]

Year	Spring wheat		Winter wheat	
	Number samples	Average 1000-kernel weight	Number samples	Average 1000-kernel weight
		g.		g.
1911	97	22.7	—	—
1912	106	25.2	47	27.6
1913	82	28.0	29	28.0

[1] Data from C. H. Bailey (1914).

TABLE V

CERTAIN PHYSICAL AND CHEMICAL CHARACTERISTICS OF THE WHEATS GROWN IN INDIA
(From the Government of India's Publication "Report on the Marketing of Wheat in India," 1937)

Province or state	Weight per bushel		Weight of 1000-kernels		Moisture		Crude protein		Crude dry gluten	
	Av.	Max.	Av.	Max.	Av.	Max.	Av.	Max.	Av.	Max.
	Lbs.	*Lbs.*	*g.*	*g.*	%	%	%	%	%	%
Punjab—General Colonies	61.5	69.0	32.3	43.0	9.83	13.19	8.78	11.12	7.76	12.13
East and Southeast	61.4	64.7	31.6	40.9	9.99	12.84	8.64	11.17	7.19	9.31
East and Southwest	61.5	65.5	34.0	42.4	9.05	10.73	8.61	9.80	8.56	9.81
North	62.2	64.8	32.7	38.7	9.89	11.78	8.17	10.94	8.30	10.12
United Provinces:										
West	61.3	65.7	32.2	45.2	10.22	12.57	8.71	10.15[1]	7.31	11.85
Central	62.7	66.2	32.8	44.4[2]	10.72	12.67	9.59	10.37	8.45	10.23
East	62.7	64.8	32.5	41.0	11.23	—	13.57[3]	—	7.02	—
U. P. Vulgare	61.2	66.3	36.8	45.6	10.44	11.66	9.24	11.12	8.30	10.35
Bombay Vulgare	62.6	64 5	37.7	44.7	10.90	12.35	10.17	13.05	11.35	14.73
Sind	60.8	65.5	31.7	39.9[4]	9.67	10.53	9.97	12.60	9.59	13.55
Bihar and Orissa	62.4	64.5	30.6	38.6	9.78	11.40	9.42	12.77	8.45	13.25
N.W.F.P.	60.6	61.7	33.5	48.0	9.64	9.86	9.40	10.20	8.11	9.60[5]
Bengal	61.4	65.7	30.6	38.6	9.26	11.35	10.79	13.51	9.47	11.49
Madras	60.6	63.3	35.7	45.6	—	—	—	—	—	—
Indian States:										
Rajputana	62.2	65.7	39.2	46.6[6]	8.90	9.22	10.48	14.65	9.26	11.88
Central India:										
Vulgare	63.2	65.3	38 5	46.2	8.86	9.55	9.63	9.83	8.45	10.32
Hyderabad	60.3	65.3	39.2	47.8	10.81	11.53	10.31	13.97	10.73	16.90
Kashmir	60.8	63.9	33.9	41.8	10.84	11.88	8.63	9.58	6.98	8.84

[1] The protein content of a sample of Cawnpore 13 was 12.31%.
[2] One sample of Cawnpore 13 weighed 50.06 g. and a durum sample 50.20 g.
[3] Only one instance.
[4] The durum samples weighed 46.55 and 48.25 g.
[5] Only three samples analyzed.
[6] Five durum samples weighed 58.25, 57.05, 53.75, 53.20, and 51.50 g.

TABLE VI

TEST WEIGHTS AND FLOUR YIELDS OF PUNJAB WHEATS

Number	Type	Irrig. or *barani*	Test weight per bushel of scoured wheat		1000-kernel weight of scoured wheat	Mottled grains	Yield of straight flour	Ratio between flour yield and bushel weight
			Winchester	Imperial				
			Lbs.	*Lbs.*	*g.*	%	%	
1	C591	Irrig.	64.7	66.8	47.3	6.6	80.0	1.23
2	C518	"	65.0	67.1	36.9	22.0	79.4	1.22
3	C518	"	65.0	67.1	34.1	21.3	78.9	1.21
4	C591	"	64.0	66.0	47.8	11.3	77.6	1.21
5	8A	"	63.5	65.5	33.8	1.3	77.1	1.21
6	8A	"	63.0	65.0	39.5	24.6	75.9	1.20
7	9D	*Barani*	64.6	66.7	38.2	56.3	68.5	1.06
8	9D	"	64.0	66.0	37.8	52.3	67.9	1.06
		HARD FEDERATION × 9D						
9	C206	Irrig.	64.0	66.0	43.5	0.7	76.3	1.17
10	C208	"	64.0	66.0	45.1	0.7	75.5	1.18
11	C209	"	63.0	65.0	37.4	0.7	72.7	1.15
12	C205	"	65.6	67.7	43.1	1.0	72.3	1.10
13	C207	"	64.0	66.0	41.0	0.7	70.8	1.10
14	C207	*Barani*	65.0	67.1	41.2	0.7	69.7	1.07
		C518 × HARD FEDERATION						
15	C210	Irrig.	63.0	65.0	48.1	8.0	75.5	1.19
16	C212	"	62.3	64.3	36.6	7.6	68.4	1.09
		C516 × C591						
17	C215	*Barani*	64.5	66.6	39.4	4.6	77.4	1.20
		HARD FEDERATION × 9D						
18	C225	Irrig.	64.0	66.0	40.9	11.6	80.0	1.25
19	C202	*Barani*	64.0	66.0	46.0	15.6	77.8	1.21
20	C229	"	64.0	66.0	40.9	4.6	75.8	1.18
21	C227	"	63.5	65.5	37.1	7.3	75.1	1.19
22	C204	Irrig.	63.3	65.3	47.1	3.0	75.5	1.19
23	C224	"	63.5	65.0	34.8	7.6	74.8	1.18
24	C203	"	63.5	65.5	45.7	24.0	74.1	1.17
25	C229	"	64.0	66.0	39.5	2.0	73.7	1.15
26	C201	*Barani*	63.6	65.6	47.0	12.6	71.9	1.13
27	C202	Irrig.	63.0	65.0	42.3	1.3	71.1	1.12
28	C228	*Barani*	64.0	66.0	44.1	14.0	70.9	1.10
29	C201	Irrig.	63.7	65.7	44.0	2.0	69.1	1.08
30	C228	*Barani*	64.5	66.6	46.2	6.3	69.0	1.07
31	C227	"	63.5	65.5	38.5	6.3	68.7	1.08

7.22 grams. The average for all Indian wheats is 34.18 g. and for the Punjab wheats 32.65 g., however.

Bailey (1914) reported the average 1000-kernel weights for some of the spring and winter wheats grown in Minnesota for the crop years 1912 and 1913. These values are given in Table IV for purpose of comparison.

Test Weight per Bushel

Test weight per bushel is an approximate measure of the plumpness of the wheat kernels. It also gives an indication of potential flour yield, since, other things being equal, plump grains usually yield more flour than shriveled grains of low weight per bushel. The test weights of the scoured wheat samples are shown in Table VI, each value recorded being an average of three determinations. It will be seen that the average of 31 samples is 63.9 lbs., with a range from 62.3 to 65.6 lbs. per Winchester bushel. Again in Table V, the average test weight of all the wheats reported is in no case less than 60.0 lbs. per bushel. The maximum value of 69.0 lbs. per bushel is reported in the instance of wheats from the canal colonies of the Punjab.

Relation between Test Weight and Flour Yield

Bailey (1924) summarized the results of two seasons' work at the Minnesota Experimental Flour Mill showing the correlation between weight per bushel and flour yield. Results by Bailey (1925) are given in the fifth table in his bulletin and indicated that flour yield generally followed weight per bushel.

Mangels and Sanderson (1925a) determined the correlation between test weights per bushel and yield of straight-run flour for seven crop years of hard red spring wheat. They found a high positive correlation and in all cases the coefficient of correlation was much higher than the probable error. The high degree of correlation is remarkable in that the data presented cover very different types of crop seasons.

A similar correlation between the test weight per bushel and flour yield was computed by the Harris formula in the instance of the data for the Punjab wheats recorded in Table VI and r was $+.219$. This means that the correlation though positive was too small to be significant. Considering the whole range of possible bushel weights for these wheats, it is seen that they are assembled at the heavy end of the range of bushel weights of wheat. The grains of all samples are almost of the same uniform plumpness and therefore there is not much variation. When the range of material studied is narrow as in the range of bushel weights used here, the variation in the flour yield must be exceedingly small if the value of r is to be large enough to be sig-

nificant, and the error in determining flour yield may thus exceed the anticipated differences.

In the study made by Mangels and Sanderson, the range in test weight per bushel for the different crop years was 22.5 lbs. while the range in flour yield was 26 lbs. In this case, however, the variation is only 3.3 lbs. per bushel.

Experimental milling.—Each sample of 2,000 grams of cleaned wheat, plus the added water for conditioning, was milled in an experimental flour mill. This mill consists of four stands of Allis-Chalmers rolls and one double stand of Wolf rolls. The break rolls have differentials of 2½:1, the smooth rolls 1½:1. Each sample was subjected to five breaks and eight reductions with bolting after every grinding operation. The temperature of the mill room was under thermostatic control and a relative humidity of 85%–90% was maintained by a Bahnson humidifier.

Flour yield.—In flour yields, Indian wheats stand high among the wheats of the world. This is supported by the figures for flour yield given by Coleman *et al.* (1930), which show that among the wheat-producing countries of the world, India stands first in yield of flour from hard red winter and soft red winter grades and second in the instance of hard red spring and white wheat grades. The flour yield ranges between 71.5% and 74.2%. Fisher and Jones (1937) point out that with careful preparation before milling a good sound sample of Karachi wheat will actually give the highest percentage of flour of all the commercial classes, this condition depending upon adequate mellowing of the wheat during conditioning, which is not easy to attain.

When we compare the flour yields of 8A, C591, and C518, now constituting by far the major portion of annual acreage under the improved wheats in the Punjab, with the flour yields of some of the world wheats as given in the analytical data of world wheats by Geddes (1937), we find that these compare very favorably.

The average flour yield (calculated on total product, not on dry wheat) of the 31 samples studied is 73.9%, which is a rather high yield of flour when the relatively simple equipment used in the experimental milling is considered. With similar equipment Bailey produced 70.2% and 69.3% of straight-grade flour from Minnesota spring and winter wheats respectively.

Eight samples of wheats C591, C518, 8A, and 9D, which are the improved wheats extensively grown at present by the Punjab farmers, yielded 75.6%. Thus C591 and C518 give a 79%–80% yield, while 8A yields 75.9% to 77% and 9D almost 68%. It is probable, however, that the 80% yield of C591 was a little too high because of the low

yield of bran, which may have been the result of under-conditioning. The second sample of C591 (serial No. 4, Table III) was therefore milled by adding 1% more water, *i.e.* 18% moisture content and the same for C518 and 8A (serial Nos. 2 and 6).

Among the new hybrids cross No. 225 gave the highest flour yield, 80%. Next was cross No. 202 (*barani*), giving 77.8%. A few others also gave a good flour yield.

Crude Protein in Punjab Wheats and Flours

The crude protein contents of wheat and flour of the samples, calculated to 13.5% moisture basis, are recorded in Table VII.

The average crude protein content in wheat of the 31 samples was 10.36%. In the first group of eight samples of C518, C591, 8A, and 9D the average crude protein was 9.3%, of which 9D had the least amount. There was a difference of 1.7% between two samples of 8A. This may have been due to the physical consistency of the two samples —one having 1.3% of mottled grains while in the second sample mottling was as high as 24.6% (Table VI).

Percentage of crude protein in the hybrid samples of wheat having Hard Federation or 9D as one of their parents was the highest, ranging from 10.6% to 12.2% with an average of 11.45%. This may have been the result of a good combination of their two parent wheats, both of which are reputed to possess good milling and baking qualities.

In the third group, *i.e.* hybrids resulting from a cross between Hard Federation and 9D, the level of crude protein was decidedly higher than in the first group. The average was 9.71% with a range of from 9.6% to 10.8%, excepting one unusual instance which was as high as 12.43%. In other words, a combination of 9D and Hard Federation has yielded hybrids which have from 1.5% to 2.7% more protein than 9D itself, which has only 8.1%.

Attention should be called to cross No. 215, a hybrid of C516 and C591. Its protein content was 9.7%, the same as that of C591. As mentioned elsewhere, C516 has very poor baking quality, and the above combination does not appear to have yielded results of any economic importance.

Reference is again made to Table V regarding the distribution of protein in wheat in different parts of India. The average ranges between 8% and 10.8% with a single instance of 13.57%. In the Punjab, the average protein content of the wheat previously reported was 8% to 9% with a maximum of 11.17%. The level of protein in the samples analyzed by us was higher, however, which suggests that a decided improvement is being and has been made on the existing stock,

TABLE VII

CRUDE PROTEIN AND CRUDE GLUTEN CONTENTS OF PUNJAB WHEATS

Type No.	Moisture content		Crude protein (N×5.7), 13.5% moisture basis		Crude gluten		
	Wheat	Flour	Wheat	Flour	Wet	Dry	Ratio
	%	%	%	%	%	%	
8A	11.2	12.1	10.65	9.57	24.7	9.4	2.62 : 1
8A	11.4	12.9	9.10	8.32	20.2	7.8	2.59 : 1
9D(B)[1]	11.3	13.1	8.06	7.35	17.9	6.8	2.63 : 1
9D(B)	11.5	13.7	8.16	7.35	18.6	6.9	2.69 : 1
C518	10.8	12.4	9.68	9.35	23.5	8.8	2.67 : 1
C518	10.8	13.3	9.63	9.10	24.7	9.0	2.74 : 1
C591	11.3	13.1	9.67	8.57	24.0	9.0	2.66 : 1
C591	11.3	13.8	9.70	8.81	24.6	8.8	2.81 : 1
			HARD FEDERATION × 9D				
C205	10.8	12.4	10.55	9.68	28.6	10.3	2.78 : 1
C206	10.8	12.8	12.19	10.83	31.3	11.2	2.79 : 1
C207	10.9	12.5	12.10	11.36	29.2	11.3	2.58 : 1
C207(B)	10.9	13.6	11.21	10.84	29.7	10.7	2.78 : 1
C208	11.7	12.8	10.89	10.10	29.0	10.6	2.74 : 1
C209	11.0	13.2	11.80	11.06	29.3	10.0	2.93 : 1
			C518 × HARD FEDERATION				
C210	11.4	12.8	10.58	9.70	27.2	9.5	2.86 : 1
C212	10.9	13.3	11.19	10.81	26.6	10.1	2.63 : 1
			C516 × C591				
C215	11.3	12.6	9.71	8.94	25.1	8.7	2.89 : 1
			HARD FEDERATION × 9D				
C201	11.1	14.5	10.70	9.95	25.1	9.6	2.61 : 1
C201(B)	11.1	13.3	10.41	9.62	24.9	9.6	2.59 : 1
C202	11.2	13.5	12.43	11.34	28.6	11.0	2.60 : 1
C202(B)	11.1	13.7	10.49	10.00	24.0	9.4	2.55 : 1
C203	11.5	12.9	10.17	9.01	24.4	9.0	2.71 : 1
C204	11.6	13.1	10.23	9.43	26.2	9.0	2.91 : 1
C224	10.9	12.4	9.80	8.93	23.5	8.7	2.70 : 1
C225	11.0	12.9	10.14	9.38	26.0	9.2	2.83 : 1
C227(B)	11.0	12.7	10.75	10.01	28.2	10.1	2.79 : 1
C227(B)	11.0	12.9	10.75	10.12	27.8	10.0	2.78 : 1
C228(B)	11.2	12.4	10.25	9.52	28.2	9.7	2.91 : 1
C228(B)	11.4	12.9	10.23	9.89	29.3	10.1	2.90 : 1
C229	11.2	12.2	9.60	8.94	30.4	9.3	3.27 : 1
C229(B)	11.8	12.4	10.38	9.85	28.9	9.9	2.92 : 1

[1] (B) denotes *barani*.

or that the crop season or the area in which these samples were grown resulted in a higher than normal protein content.

When we compare these wheats with hard bread wheats grown in other parts of the world, we find that as regards percentage of crude protein these wheats, with an average of 10.36%, are in the medium-to-low range. It is of the same order of magnitude as the softer types of

wheats reported in the tri-state (Ohio, Indiana, and Michigan) surveys by Bayfield (1933) which ranged as follows:

Year	Average Crude Protein
	%
1931	10.67
1932	9.80
1933	11.26
1934	11.64
1935	9.88
1936	9.64

Crude Protein in Wheat and Physical Texture of the Wheat Kernel

Grain texture, as determined by the percentage of vitreous, mottled, and soft grains in a wheat sample, bears a certain relation to protein content and consequently to baking strength. The physical texture of the grain of the samples is recorded in Table VI, and the protein content in Table VII. Samples of 9D have the highest percentage of mottled grains and the lowest of protein content, while C205–208 all have mottled grains less than 1%, and the percentage of crude protein is proportionately higher, perhaps highest. Coleman, Fellows, and Dixon (1925) have also shown that dark, hard, and vitreous kernels have more protein than the soft and chalky ones.

This relation between kernel texture and protein in wheat has been statistically examined by Mangels and Sanderson (1925b), Hayes, Immer, and Bailey (1929), Singh (1935), and Aamodt and Torrie (1935). The conclusion arrived at by these workers is that there exists a significant positive correlation between the vitreousness of the kernels and the protein in wheat.

A similar correlation was worked out in this case, with the difference that in this case the percentage of mottled grains was correlated with the percentage of protein in wheat. The correlation has been found to be significant but negative, indicating that mottling of grain is inversely proportional to the protein content ($r = -.66 \pm .0675$).

Crude Gluten

Crude gluten in the experimentally milled flours was determined by the A. A. C. C. method (1935). The percentage of, and the ratio between, wet and dry gluten (dried to constant weight in an air oven at 97° to 99° C.) is shown in Table VII. The results corroborate the statement, made earlier, that the percentage of dried crude gluten is approximately equivalent to the percentage of crude protein in flour.

Diastatic Activity

The determination of diastatic activity of the flour samples shown in Table VIII was made by the Blish and Sandstedt (1933) method as

TABLE VIII

DIASTATIC ACTIVITY
(Maltose produced in 10 g. flour in one hour)

Type	Maltose	Type	Maltose	Type	Maltose
	mg.		*mg.*		*mg.*
C591	289	Hard Federation × 9C		Hard Federation × 9D	
C518	286	C208	276	C204	291
8A	283	C209	268	C229	259
9D	274	C207	237	C225(B)[1]	258
				C228	257
C591	271	C206	230	C224	256
C518	269	C205	221	C203	254
9D	266			C201	253
8A	258	C518 × Hard Federation		C202	241
		C210	272	C202(B)	235
				C202	235
		C516 × C591		C229(B)	231
		C215	238		

[1] (B) denotes *barani*.

described in A. A. C. C. Cereal Laboratory Methods (1935). It will be seen that the diastatic activity of these samples seems to have been fairly high, ranging between 221 and 291 with an average of 256.7, with the exception of two or three cases. The range of variation was not great, however. The diastatic activity of the C591, C518, 8A and 9D wheat group was high; average of these eight samples was 274.5. Next comes the group of hybrids, having Hard Federation and 9D as parents, with an average of 251.7, while among the progeny of Hard Federation and 9C it was 246.7.

Chapati-Making Quality

The bulk of the Punjab wheat is consumed in the form of *chapati* (unleavened pan-baked cakes). Generally the quality of *chapati* can be judged by two factors: (1) its softness and flexibility, which may perhaps be due to the quantity and quality of protein in the flour, and (2) its sweet and flavory taste which may be due to the amount of sugar in the flour plus the hydrolysis of starch into sugar by the diastatic enzymes as a result of addition of water to the flour to make dough. Therefore a wheat in order to be of good *chapati*-baking value should have, no doubt, a fairly high diastatic activity.

Of the wheats listed in the upper group (Table IX), C591 and 8A make very good *chapaties* of excellent flavor, C591 perhaps the better, and C518 almost as good as 8A. Good *chapaties* cannot be made from 9D, however. This may perhaps be due to less protein in the flour. Of the 31 samples analyzed, 9D has been shown to possess the least amount of protein both in the wheat and the flour.

Another point in the testing of the quality of *chapati* is its keeping quality. The common practice is to bake them fresh for every meal. A soft silky *chapati* on keeping, say overnight, will not have its softness impaired or lessened to a very great extent. This may be due to the water-holding capacity of protein.

Sufficient water is added to the flour to make it into a fairly stiff dough. This dough consists of flour and water only. It is kneaded with the hand and allowed to stand for an hour or so, so that the moisture absorption is complete and the dough assumes a definite shape and consistency. It is kneaded again for a short time before baking.

The dough then is divided into small dough balls. Each dough ball is rolled and flattened and made into a *chapati*, which is then put on the hot plate. After a minute or two the side of the *chapati* in contact with the hot plate becomes stiff as a result of dehydration of protein and starch. It is then turned over so that the other side gets stiff likewise. It is then either turned over again on the hot plate or put on the glowing charcoal, where it puffs and becomes ready for use. It is usually greased with butter before eating and is always eaten when it is fairly warm.

Relation Between Diastatic Activity and the Protein Content of Flour

If we examine the diastatic activity and the protein content of these flours, we find that the averages of these two variables are as follows:

Group	Average of	Diastatic Activity	Protein in Flour
		mg.	%
Duplicate samples of C518, C591, 8A, and 9D	8 samples	274.5	8.55
Hybrid (H.F. × 9D)	11 samples	251.7	9.58
Hybrid (H.F. × 9C)	5 samples	246.4	10.64

The fact, as suggested by the above data, that the diastatic activity appears to have an inverse relation with the protein content of flour warranted statistical examination of the case. The coefficient of correlation between these two variables was, therefore, computed with the following result: $r = -.422 \pm .1067$.

This shows that as far as these data are concerned there was a small negative correlation with a large probable error, however.

While diastatic activity is not directly associated with, or due to the protein content of, wheat, it apparently occurs not uncommonly that certain factors of environment during the late growth period of the plant influence both variables concurrently. Thus arid or drought conditions tend to result in a high-protein wheat grain. The same conditions also seem to result in a high degree of biological maturation of the grain which implies a low enzymic activity in general. Thus the

diastatic activity tends toward a minimum when the protein content is approaching an abnormally high level.

A natural corrollary of these conditions is a tendency towards dry, well-cured wheat under drought conditions, which implies a minimum of field-damaged and sprouted kernels. This also maintains a minimum of diastatic activity in the grain.

Determination of the Gassing Power of Flour

In order to study the relation between the diastatic activity and gassing power, four flours of varying diastatic activity were selected. The method employed was the same as adopted by Bailey and Johnson (1924). In terms of volume of gas production and dough expansion, C591 gave the maximum values, C518 was second, C229 was third, and C201 was lowest, which is also the order of their diastatic activity. To what extent the diastatic activity is influenced by the environment in which wheat is grown can be judged by the fact that the wheat C229 when grown under rain-dependent (*barani*) conditions showed a diastatic activity of 254 and the same wheat under the irrigated condition gave only 176, a difference of 78 units. Similar observations were made by Mangels (1926) and Swanson (1935).

Water Absorption

Water absorptions of the flour samples under investigation were determined by the farinograph except in some cases where this had to be done by hand kneading of 25 g. of flour, in duplicates, by the addition of necessary amounts of distilled water to make dough of a workable consistency. The amount of water used multiplied by four is recorded in the case of hand-kneading tests.

The water absorptions are given in Table IX. It is evident that these flours have a very high water absorption, the average of 31 samples being 72.6%. This is all the more remarkable in view of the average protein in the flour, which was only 9.60% and considerably lower than the bread wheats of United States, Canada, and Russia, etc. What this absorption may be due to cannot be explained at present. This is a problem requiring further elucidation.

Reference is made to the summary of milling and baking qualities of the world wheats, Coleman *et al.* (1930), page 218. It will be seen that Indian wheats belonging to the predominating classes of hard red winter and white stand very conspicuously at the top in water absorption. In this respect they compare still more favorably with the world wheats (Geddes, 1937). Fisher and Jones (1937), page 95, also found the water absorption of Karachi wheat to be relatively very high with a correspondingly high bread yield.

TABLE IX

WATER ABSORPTIONS AND BAKING RESULTS

Serial No.	Type	Water absorption			Two min. mixing, 1 hr. fermentation		One min. mixing, 1½ hrs. fermentation		Change in bread properties from 1st to 2nd baking method
		By farinograph	Used in baking	Difference	Loaf vol.	Baking score	Loaf vol.	Baking score	
		%	%	%	cc.		cc.		
1	8A	72.0	72.0	+0.0	510	56.0	515	53.5	0
2	8A	70.6	73.0	+2.4	440	45.0	475	53.5	+
3	9D	70.6	75.0	+4.4	485	—	452	51.2	−
4	9D	68.0	75.0	+7.0	525	60.5	480	54.0	−
5	C518	76.8	79.0	+2.2	455	46.5	495	54.5	+
6	C518	72.0	74.5	+2.5	465	50.5	500	51.0	+
7	C591	77.3	81.0	+3.7	485	—	403	36.3	−
8	C591	68.6	72.0	+3.4	500	55.0	500	50.0	0
	C516 × C591								
9	C215	66.8	70.0	+3.2	515	63.5	455	52.5	−
	HARD FEDERATION × 9C								
10	C205	68.6	70.0	+1.4	530	59.0	525	60.5	0
11	C206	69.3	71.0	+1.7	490	52.0	570	67.0	++
12	C207	68.3	72.0	+3.7	485	50.5	613	74.3	++
13	C207B	67.0	68.5	+1.5	605	69.5	615	68.5	+
14	C208	75.0*	75.0	+0.0	535	63.5	538	58.8	0
15	C209	76.0	77.0	+1.0	580	72.0	425	44.5	−−
	C518 × HARD FEDERATION								
16	C210	78.0*	77.0	−1.0	550	67.0	518	58.8	−
17	C212	66.6	69.0	+2.4	515	55.5	500	54.0	−
	HARD FEDERATION × 9D								
18	C201	64.4*	65.0	+0.6	515	50.5	508	53.8	0
19	C201B	71.3	73.0	+1.7	510	53.0	485	49.5	−
20	C202	70.1	70.1	+0.0	440	46.0	465	43.5	+
21	C202B	68.4	71.0	+2.6	505	53.5	470	47.0	−
22	C203	71.0*	71.5	+0.5	440	45.0	445	45.5	0
23	C204	75.2	76.0	+0.8	500	53.0	493	55.3	0
24	C224	69.6	74.0	+4.4	455	50.5	525	60.5	++
25	C225	70.5	74.0	+3.5	575	69.5	525	62.0	−
26	C227	67.8	69.0	+1.2	500	58.0	545	63.5	+
27	C227	68.0*	71.0	+3.0	550	67.0	535	62.5	−
28	C228	73.3	75.0	+1.7	535	61.5	508	57.8	−
29	C228	70.0*	72.0	+2.0	580	70.0	500	55.0	−
30	C229	67.2*	67.5	+0.3	465	54.5	500	58.0	+
31	C229	71.3	73.0	+1.7	485	57.5	480	58.0	0

* Determined by hand kneading.

Protein Content and Water-absorbing Capacity of Flour

It is generally thought that high-protein flours have higher water-absorbing capacity and *vice versa*, although this is not invariably the case, as considerable variation in absorption may be encountered in wheat flours of the same protein content but milled from different lots.

Mangels (1928) worked out coefficients of correlation for four crop years. He found positive correlations for three years between +.2 and +.3 and a negative correlation in one year. He felt that coefficients of this magnitude do not indicate a high degree of correlation between protein content and water absorption.

In order to ascertain whether any relationship exists between the protein content and water absorption in the instance of the flours involved in these studies, the coefficient of correlation of these two variables was computed and found to be $r = +.6747 \pm .066$. This shows that so far as these data are concerned there is a high positive correlation.

Baking Tests

For experimental baking test the standard test formula of the American Association of Cereal Chemists was followed except that the sugar was doubled. The formula used was as follows:

Flour 100 g. (100%) at 15% moisture
Yeast 3 g. (3%)
Salt 1 g. (1%)
Sugar 5.0 g. (5%)

In the light of information yielded by the farinograph, that the dough formation of these flours took less time and the range of minimum dough mobility was rather narrow, it was thought advisable not to subject them to a long high-speed mixing or long fermentation. The low range of protein in these flours was another factor in favor of short fermentation. Accordingly a test baking, with two minutes of mixing and one hour of fermentation, was conducted. The loaf-volume figure in each case is a measurement of a single loaf, and accordingly too much reliance cannot be placed on these results. Nevertheless they give some indication of baking quality when they are compared with the results of the second baking.

It had been observed in this laboratory that the amount of water added to certain types of flour for bread making is generally 1% to 2% less than determined by the farinograph when titrated to 500 Brabender units. The same thing was done in this case, but it was found that the dough was quite stiff. Water was then added equal to that found by the farinograph and the dough still felt too stiff. Finally 1% to 2%

more water than the farinograph determination was given. The second baking was done by the addition of 1% to 4% more water; in some cases even more. Low loaf volumes with sample Nos. 10, 11, 12, 24, and 26 in Table IX very probably are due to insufficient water for optimum dough consistency.

In a second series of baking tests the mixing time was reduced from two minutes to one minute and the fermentation period was increased from 1 hour to 1½ hours. The results are shown in Table IX. Each figure is an average of two loaf-volume measurements. This is almost of the order of American soft winter wheats as reported by Bayfield (1933).

There is, however, one factor to be taken into consideration and that is that this average of 502 cc. is the result of great variations in the loaf volume due, perhaps, to the different genetical composition of the hybrids. When the selection is made amongst those which have higher loaf volume as well as other desirable characteristics much more favorable results are apparent.

If we compare the loaf volumes of these flours yielded by two different baking methods (Table IX), we find that there were some which gave increased loaf volume when mixing time was decreased to 1 minute and fermentation time increased to 1½ hours. There were others which did not respond to the changed treatment, and again there were some which gave decreased loaf volumes. Where the change from the first to second treatment was negligible it is indicated by zero. In the remaining cases the magnitude of such a change is expressed by the number of (+) or (−) signs for the positive and negative change respectively. On the whole, crosses 205, 206, 207 (Hard Federation × 9C) and crosses 224, 225 and 227 (Hard Federation × 9D) have given all around good results.

Viscosities of Flour-in-Water Suspensions

The standard A. A. C. C. method detailed in Cereal Laboratory Methods (1935) was employed. A MacMichael viscosimeter with a No. 30 wire disk bob and speed of 12 rpm. was used. All the determinations were made at room temperature (80°F.). The suspensions were prepared without previous removal of electrolytes, and they were acidulated with lactic acid. The results reported in Table X are averages of duplicate determinations of 24 selected samples. These results show two sets of conditions: First, where the maximum viscosity occurs at the point when the first cubic centimeter of lactic acid is added. The second addition of 2 cc. decreased the viscosity a great deal. The additions of subsequent third and fourth 2-cc. portions

TABLE X

VISCOSITIES OF FLOUR-IN-WATER SUSPENSIONS

Type	Viscosity, degrees MacMichael					Increase or decrease (−) in viscosity from added acid (°MacM.)				Type of viscosity
	Acid used, cc.					Acid used, cc.				
	0	1	3	5	7	1	3	5	7	
C518	11.15	44.5	10.5	12.5	17.5	33.0	−34.0	2.0	5.0	A+1
C518	9.0	39.5	8.5	12.0	16.5	30.5	−31.0	3.5	4.5	A+1
C591	11.0	50.5	13.5	11.0	13.0	39.5	−37.0	−1.5	2.0	A+2
C591	7.5	41.5	8.0	9.5	13.0	34.0	−33.5	1.5	3.5	A+1
8A	9.0	25.0	10.0	15.5	21.0	16.0	−15.0	5.5	5.5	A
8A	10.5	25.5	11.0	10.0	13.0	15.0	−14.5	−1.0	3.0	A
9D	11.0	21.5	17.5	26.0	32.0	10.5	− 4.0	8.5	6.0	B
9D	9.0	16.5	13.0	22.5	27.5	7.5	− 3.5	9.5	5.5	B
C518 × HARD FEDERATION										
C210	9.0	29.0	11.0	20.0	26.5	20.0	−18.0	9.0	6.5	A
C212	7.5	20.0	11.5	20.5	27.0	12.5	− 8.5	9.0	6.5	B
C516 × C591										
C215	8.5	19.0	17.0	27.0	32.0	10.5	− 2.0	10.0	5.0	B
HARD FEDERATION × 9C										
C205	6.5	10.0	25.5	38.0	42.0	4.0	15.5	12.8	4.0	B+1
C206	8.0	12.0	30.0	47.0	52.0	4.0	18.0	17.0	5.0	B+2
C207	5.0	8.5	38.5	46.0	50.0	3.5	30.0	7.5	4.0	B+2
C209	10.0	33.0	10.5	14.5	19.5	23.0	−22.5	4.0	5.0	A
HARD FEDERATION × 9D										
C201	5.0	8.0	20.5	26.5	29.5	3.0	12.0	6.0	3.0	B
C201	8.0	14.5	20.5	33.5	39.6	6.5	6.0	13.0	6.0	B
C202	7.5	13.0	15.5	25.5	31.5	5.5	2.5	9.5	6.5	B
C203	7.5	15.5	12.5	23.5	28.0	8.0	− 3.0	11.5	4.5	B
C204	11.0	18.0	30.5	49.5	52.5	7.0	12.5	19.0	3.0	B+2
C224	9.0	10.0	31.5	44.0	51.5	1.5	21.0	12.5	7.5	B+2
C225	9.0	12.0	31.0	43.5	51.0	3.0	19.0	12.5	8.0	B+2
C227	11.0	13.0	26.5	40.0	42.0	2.0	13.5	13.5	2.0	B+1
C228	8.0	12.5	30.0	43.5	49.0	4.5	17.5	13.5	5.5	B+2
Amer. wheat flour	8.0	10.5	41.5	63.5	71.0	2.5	31.5	21.5	8.0	B+2

tended to increase the viscosity but not to an appreciable extent. In the second case, however, the increase in viscosity was approximately proportional to the successive increments of the acid and the maximum viscosity occurred on the addition of 7 cc. of acid.

On the basis of the above distinctions these wheat samples have been classified according to the type of viscosity exhibited by each. The first case, where the maximum viscosity occurred with the addition of 1 cc. of acid, has been designated by *A*, *A+1*, and *A+2*, depending upon the extent of increase, *A* representing the lowest maximum and *A+2* the highest maximum.

The other condition (with 7 cc. of acid) is represented by *B*, *B+1*, and *B+2*, *B* representing the lowest maximum and *B+2* the highest maximum. The arbitrary limits have been fixed as follows:

A	Less than 35°	MacMichael by the addition of 1 cc. acid
A+1	Between 35°–45°	MacMichael by the addition of 1 cc. acid
A+2	More than 45°	MacMichael by the addition of 1 cc. acid
B	Less than 35°	MacMichael by the addition of 7 cc. acid
B+1	Between 35°–45°	MacMichael by the addition of 7 cc. acid
B+2	More than 45°	MacMichael by the addition of 7 cc. acid

According to the two divisions mentioned above into which these wheats have resolved themselves as regards viscosity behavior two things become apparent:

(1) C518, C591, 8A and 9D exhibit viscosities of the *A* type. The change in viscosity is very great in the first two, less in 8A, and least in 9D. C215, whose parents both happened to be the indigenous wheat hybrid selections, behaved in like manner.

(2) In the case of hybrids resulting from a combination of the local wheat selections with Hard Federation, a foreign bread wheat, the viscosity behavior in the majority of cases is shown to be analogous to the bread wheats of the United States or Canada, for example, though the maximum viscosity is less than these, perhaps because of less crude protein in wheat and flour.

So far as the literature is concerned no case has been recorded where the maximum viscosity occurred by the addition of first 1 cc. of lactic acid and then the following addition of 2 cc. acid decreased the viscosity. The reason for this is not apparent and constitutes a problem requiring further research. This behavior might be suspected to be a function of the hydrogen-ion concentration of the preparations, pH of the flour-in-water suspensions having been shown previously to influence the viscosity. When pH was determined the problem was not clarified, save that in C518 and C591 the maximum viscosity occurred at pH 4.5 while in the other case the maximum viscosity occurred at pH 2.8 to 3.0. The viscosity measurements in °MacM and

the relative pH of the four typical flours with different increments of acid are shown in Table XI.

TABLE XI

VISCOSITIES AND pH DETERMINATIONS

Treatment 1*N* lactic acid	American wheat		C206		C518		C591	
	pH	Viscosity	pH	Viscosity	pH	Viscosity	pH	Viscosity
cc.		°*MacM.*		°*MacM.*		°*MacM.*		°*MacM.*
–	5.78	8.0	5.78	8.0	5.98	11.5	5.93	7.5
1	4.41	10.5	4.38	12.0	4.56	44.5	4.51	41.5
3	3.58	41.5	3.58	30.0	3.77	10.5	3.80	8.0
5	3.18	63.0	3.07	47.0	3.38	12.5	3.42	9.5
7	2.98	71.0	2.82	52.0	2.77	17.5	2.81	13.0

C518 and C591, viscosity *A* type; American wheat and C206, viscosity *B* type.

Carotenoid Pigments

The method described by Ferrari and Bailey (1929) was used in the extraction and estimation of the carotenoid pigments and the results for the different flours examined are recorded in Table XII as the average of two observations. The range of carotenoid pigment content was from 2.50 to 1.42 parts per million. The flours milled from wheats on the approved list, 8A, 9D, C518, and C591, have a carotenoid content of 2.05 or above, while there is some variation among the new hybrids.

TABLE XII

CAROTENOID PIGMENT CONTENT OF DIFFERENT WHEAT SAMPLES

Serial No.	Type or cross No.	Parentage	Carotenoids
			ppm.
1	8A	——	2.50
2	C207	H.F. × 9C	2.33
3	C591	——	2.27
4	9D	——	2.23
5	C212	C518 × H.F.	2.15
6	C518	——	2.05
7	C215	C516 × C591	2.00
8	C205	H.F. × 9C	1.90
9	C206	H.F. × 9C	1.80
10	C225	H.F. × 9D	1.80
11	C202	H.F. × 9D	1.77
12	C202B	H.F. × 9D	1.75
13	C208	H.F. × 9C	1.75
14	C225	H.F. × 9D	1.68
15	C228	H.F. × 9D	1.62
16	C201	H.F. × 9D	1.55
17	C209	H.F. × 9C	1.55
18	C210	C518 × H.F.	1.54
19	C203	H.F. × 9D	1.47
20	C204	H.F. × 9D	1.45
21	C229	H.F. × 9D	1.42

With the exception of C207 and C212 the rest have a carotenoid content less than 2.0 and as low as 1.42 ppm. It is possible that this may be due to less pigment in Hard Federation, which is one of the parents of these hybrids.

If we compare the range of the carotenoid pigment content of these wheats with wheats of the world, we find that they compare favorably. Worzella and Cutler (1935) have presented (page 710) the results of carotenoid contents of 29 soft and semi-hard American wheat varieties for the crop year 1932–33 and 1933–34, which ranged from 1.70 to 3.80 in 1933, and 1.80 to 3.80 in 1934 with an average of 2.37 and 2.41 ppm., respectively.

Whiteside, Binnington, and Geddes (1933) have given carotene values of gasoline extracts of straight-grade flours of Marquis and Garnet wheats grown at 14 different places in Western Canada in 1930. For Marquis the carotene values ranged between 1.44 and 2.69 with an average of 1.97 and for Garnet between 1.79 and 3.86 with an average of 2.45 ppm.

Farinograph Study

The farinograph curves of some of the flour samples are shown in Figure 4. It will be observed that in case of C591, C518, 8A, and 9D

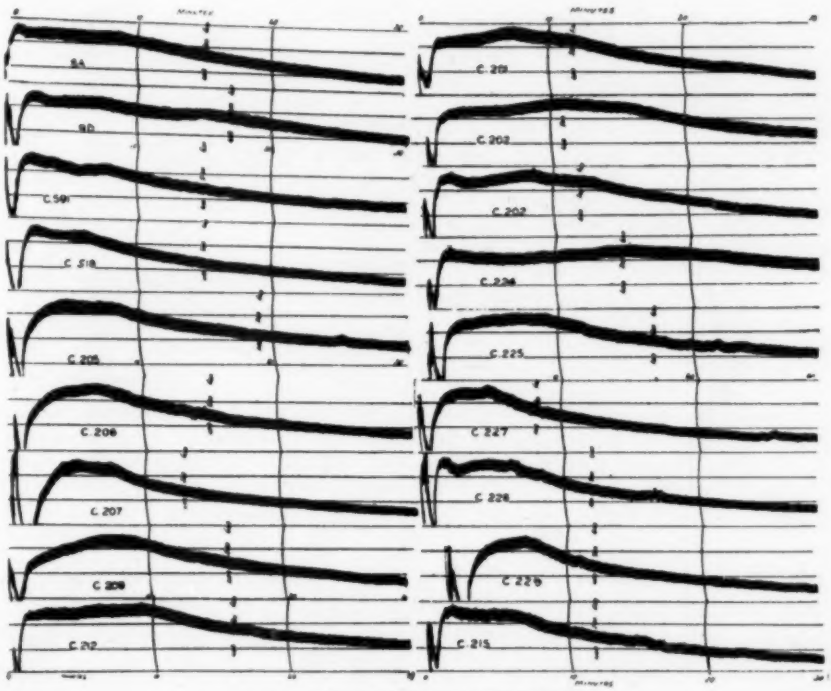

Fig. 4. Farinograph curves of Punjab flours.

the time required for dough development was rather short. This is a characteristic of many typical soft wheats. The dough mixing stabilities of C591 and C518 were lower than were those of 8A and 9D. Although the protein content in 9D was least of all, the nature of the curve suggests a good quality of protein. C215 (C516 × C591) behaved almost exactly like C591, neither of which appeared to possess a good quality of protein.

Some of the hybrids resulting from the crossing of Hard Federation × 9C and Hard Federation × 9D possess relatively superior colloidal dough properties so far as can be determined by farinograph curves. An all-round improvement seems to have been accomplished by hybridizing yet the dough-mixing stability is much less than for bread wheats of the United States and Canada. Cross 224 (Hard Federation × 9D) showed a distinct improvement over 9D in both quality and quantity of protein, while C205, C206, C207, C224, and C225 gave good baking results as well.

Quantitative Determination of the Degree of Dough Softening

Softening can be well illustrated by a reference to a farinogram in Figure 5. The maximum plasticity (minimum mobility) of the dough

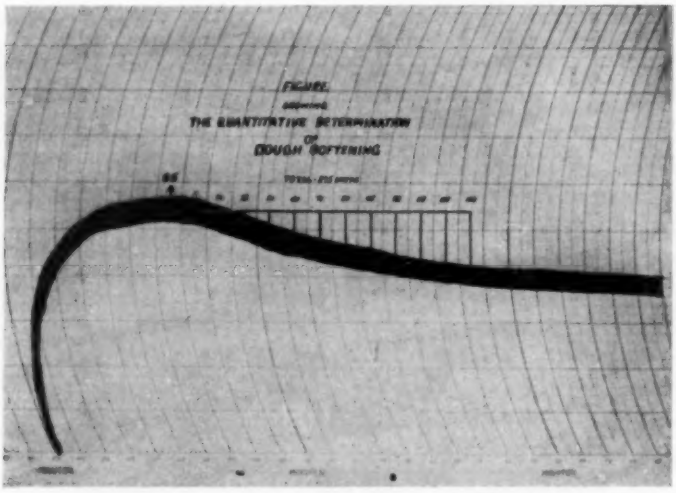

Fig. 5. Quantitative measurement of dough softening.

appears to be attained at the end of the sixth minute and begins to decline after 6.5 minutes. Just where the curve begins to fall, a point is marked and every minute thereafter up to twelve minutes. A horizontal line is projected from the first point and the distance between the

points marked on the curve and the parallel line is measured and expressed in terms of farinograph units. In this case the value of the first distance is 5, of the second 20, and so on up to the twelfth minute. All these values are then summated. The sum (895 units in this case) expresses the amount of the softening of the dough. The abrupt or gentle fall in the curve indicates the degrees of softening. These values are comparative only and give an indication of the degree of resistance to mixing which a dough offers to the mechanical abuse to which it is subjected. The summated values of the extent of dough softening of the flour samples are given in Table XIII.

TABLE XIII

RESULTS OF FARINOGRAPHIC STUDY WITH WHEAT FLOUR SAMPLES

Type	Max. dough development		Departure from minimum mobility (softening) at end of successive minutes (1–12)												Summation of 12 "softening" values
			1	2	3	4	5	6	7	8	9	10	11	12	
	Units	*min.*	*Units*												
8A	550	1.25	15	15	20	22	22	25	24	30	38	50	60	65	386
9D	540	2.5	20	20	20	20	20	28	40	46	50	50	50	50	414
C518	550	2.5	18	20	20	23	40	60	75	85	95	105	110	120	771
C591	550	2.0	18	22	35	45	40	40	50	60	70	90	100	100	670
C516 × C591															
C215	550	2.25	10	15	10	15	20	50	65	80	95	95	110	110	675
HARD FEDERATION × 9C															
C205	530	5.0	5	6	7	18	30	45	60	65	75	80	90	95	576
C206	540	6.5	5	20	35	50	60	70	80	95	110	115	125	130	895
C207	545	4.5	0	0	5	25	45	60	75	95	105	115	120	130	775
C207(B)	540	5.0	5	20	40	50	68	80	90	100	110	120	123	130	936
C518 × HARD FEDERATION															
C212	550	3.0	5	5	0	0	0	0	0	0	10	20	50	55	145
HARD FEDERATION × 9D															
C201	520	2.0	0	0	0	0	0	0	0	0	0	0	10	25	35
C202	530	3.0	0	0	0	0	0	0	0	0	0	0	0	0	0
C202(B)	550	2.5	20	15	10	0	0	0	0	5	10	15	30	40	145
C224	530	2.25	10	20	20	25	25	20	15	10	10	5	0	0	160
C225	555	7.0	0	4	15	25	45	60	70	75	80	90	100	105	669
C227	540	4.0	0	10	30	55	70	80	90	100	110	120	125	135	925
C228	550	2.5	20	5	0	5	20	35	50	70	80	95	105	110	595
C229	540	6.0	25	45	70	80	95	110	120	130	135	140	145	150	1245

Brabender's Single-Figure Evaluation of Farinograms

Recently Brabender (1937) has formulated a method with which the farinogram of a flour dough is evaluated by a single-figure score. This consists in combining the three variables, *viz.* the time in minutes of maximum dough development, degree of dough softening in units, and the single-figure score, in a three dimensional chart. First the difference between maximum consistency and consistency after 12 minutes of mixing is noted. This difference is matched on the chart against the time required to reach maximum consistency. The relative single-figure score is then directly read on the chart. A high score is presumed to imply superior quality. The results of evaluating the curves by the single-figure-score method are given in Table XIV.

TABLE XIV

SINGLE-FIGURE SCORES

Type	Max. dough development		Fall after 12 min.	Difference	Single-figure score
	Min.	*Units*	*Units*	*Units*	
8A	1.25	550	485	65	47.0
9D	2.5	540	490	50	53.5
C518	2.5	550	430	120	41.3
C591	2.0	550	440	100	42.8
			C516 × C591		
C215	2.5	550	440	110	42.0
			HARD FEDERATION × 9C		
C205	5.0	530	435	95	55.5
C206	6.5	540	410	130	60.0
C207	4.5	545	415	130	50.0
C207(B)[1]	5.0	540	410	130	52.5
			C518 × HARD FEDERATION		
C212	3.0	550	495	55	53.5
			HARD FEDERATION × 9D		
C201	2.0	520	495	25	58.5
C202	3.5	530	530	0	64.0
C202(B)	2.5	550	510	40	55.4
C224	2.25	530	530	0	62.0
C225	7.0	555	450	105	63.0
C227	4.0	540	405	135	47.0
C228	2.5	550	440	110	43.0
C229	6.0	540	390	150	56.0

[1] (B) denotes *barani*.

Dough Softening and Single-Figure Scoring of Farinograms

In Table XV are the summated values giving the extent of dough softening as a result of the breaking down of dough structure in 12

minutes after the minimum dough mobility was recorded. In the right-hand column are the results of single-figure evaluations of the farinograms. These results do not exhibit any definite relationship. The coefficient of correlation between these two variables was computed and the value of $r = -.035 \pm .157$, which is an insignificant coefficient.

TABLE XV

DATA REGARDING DOUGH SOFTENING AND SINGLE-FIGURE SCORING OF THE FARINOGRAMS

Serial No.	Type	Summated values[1]	Type	Single-figure score[2]
		Units		
1	C202	0	C202	64.0
2	C201	35	C225	63.0
3	C202(B)[3]	145	C224	62.3
4	C212	145	C206	60.0
5	C224	160	C201	58.5
6	8A	186	C229	56.0
7	9D	414	C205	55.5
8	C205	576	C202(B)	55.4
9	C228	595	9D	53.5
10	C225	669	C212	53.5
11	C591	670	C207(B)	52.5
12	C215	675	C207	50.0
13	C518	771	8A	47.0
14	C207	775	C227	47.0
15	C206	895	C228	43.0
16	C227	925	C591	52.8
17	C207(B)	936	C215	42.0
18	C229	1245	C518	41.3

[1] Arranged in ascending order. [2] Arranged in descending order. [3] (B) denotes *barani*.

Summary

Results of a biochemical and technological study of some Punjab wheat varieties grown on the Botanical Farm of the Punjab Agricultural Research Institute, Lyallpur, India, in 1935–36 are reported. Thirty-one samples were used in this study, of which 8 samples (duplicates) 8A, 9D, C518, and C591, are improved wheat types, on the approved list of the Punjab Department of Agriculture and extensively grown by the farmers. Out of a total acreage of 9 to 10 million acres annually cropped to wheat, these four types occupied as much as 50%, and 8A alone occupied 3,161,700 acres. The remaining 23 samples are all recent hybrids selected from the cross-bred material. These are still in the experimental stage. The purpose of this investigation was to determine their milling and baking qualities according to the fermented-bread-making standards and methods.

The average 1000-kernel weight of these 31 samples was 41.4 g. with a range of between 33.8 and 48.1 g., which indicates plump, heavy

kernels and moderate variation in the kernel weights. The average of these samples exceeds the average value for all India wheats by 7.22 grams.

The average test weight per Winchester bushel of these samples was 63.9 lbs. with a range from 62.3 to 65.6 lbs.

Indian wheats as a rule are classed as dry wheats, since the period during which they are harvested and threshed is a very hot and dry one. Coleman *et al.* (1930) give moisture data between 9% and 10% for some of the Punjab wheats examined. The moisture contents of samples reported here were a little higher, ranging between 10.8% and 11.8%. The small variation may be due to the fact that wheats tend to absorb moisture in storage during the monsoon (rainy) period.

In flour yields, Indian wheats stand high among the wheats of the world. This is supported by the data of Coleman *et al.* (1930) and Geddes (1937). The average flour yield of the 31 samples tested was 73.9% and that of the samples of C518, C591, 9D and 8A was 75.6%.

Among the new hybrids C225 gave the highest flour yield, 80%, and the next in order was C202 (*barani*), yielding 77.8% flour.

The coefficient of correlation computed between the bushel weight and flour yield was +.219. The correlation, though positive, is too small to be significant, which may have been due to the uniformly high bushel weights of these wheats. So far as crude protein in wheat was concerned, these wheats fell within medium-to-low range with an average of 10.36%. The group containing 8A, 9D, C518, and C591 averaged 9.3%. In the group of hybrids with Hard Federation × 9C as parents the average crude protein was 11.45%, ranging from 10.6% to 12.2%. This may have been due to the combination of wheats both of which are reputed to possess good milling and baking qualities.

The average crude protein of all India wheats ranges between 8% and 10.8%, that previously reported for the Punjab being between 8% and 9%. The level in these samples was, however, higher, which shows an improvement on the existing stock. The coefficient of correlation was computed between crude protein in wheat and the percentage of mottled grains in each sample, with $r = -.66 \pm .06775$. Crude gluten generally equals the crude protein in flour.

Diastatic activity ranged between 221 and 291, with an average of 256.7. With the exception of two or three cases the range of variation was not very large. The average of the group of approved wheat varieties (8A, 9D, C518 and C591) was 274.5. Compared with data for world wheats reported by Geddes, these wheats are high in diastatic activity.

The bulk of the Punjab wheat is consumed in the form of *chapaties* (unleavened pan-baked cakes). Therefore the method of *chapati* baking and testing is described.

The water absorption of flours milled from these wheats averaged 72.6%. This is very high indeed in view of the low average protein content.

A high positive correlation between the water absorption of flour and protein content value was observed, $r = +.6747 \pm .066$. Mangels found a low positive correlation, between 0.2 and 0.3, which did not indicate a high degree of correlation between these two variables in the samples tested by him.

The average loaf volume was 502 cc. with a maximum of 615 cc., which is of the order of American soft winter wheats. There is one factor to be taken into consideration, however, that this average was the result of a great variation in the loaf volume due perhaps to the different genetical make-up of the hybrids. On the whole crosses 205, 206, 207 (Hard Federation × 9C) and crosses 226, 225, and 227 (Hard Federation × 9D) gave good baking results.

The results of viscosity determinations show that among these wheats there existed two sets of conditions: the first, or *A* type, where the maximum viscosity occurred at the point when the first cubic centimeter of lactic acid was added, the second addition (2 cc. acid) decreased the viscosity substantially, while a third and fourth addition of 2 cc. acid each tended to raise the viscosity, though not to a substantial extent. In the second case, or *B* type, the increase in viscosity was fairly regular with successive increments of the acid and the maximum viscosity occurred on the addition of 7 cc. of acid. Indigenous selections almost all show *A* type, while hybrids resulting from a cross where one of the parents is Hard Federation exhibit the *B* type of viscosity. The literature does not record instances where the maximum viscosity occurred with the addition of the first cubic centimeter of lactic acid and the following addition of 2 cc. of acid decreased the viscosity.

The pH of each system was determined, and in the *A* type the maximum viscosity occurred at pH 4.5 while in the *B* type the maximum viscosity occurred at pH 2.8 to 3.0.

Carotenoid pigment of the flours ranged from 2.50 to 1.42 parts per million. Samples of 8A, 9D, C518, and C591 had a carotenoid content above 2.05 ppm., and there was some variation among the new hybrids.

Farinograms show that the dough development times of these flours with few exceptions were short. The range of dough stability was also comparatively narrow. Some of the hybrids of Hard Federation × 9C

and Hard Federation × 9D showed better dough properties, however, so far as was evident from their farinograms. Evaluation of the farinograms was made by two methods, (1) the summation of the decrease in consistency at intervals of one minute for 12 successive minutes after the maximum consistency or minimum mobility was reached, and (2) Brabender's new method of single-figure scoring of farinograms. No relation between these two evaluations appeared in these instances ($r = -.035 \pm .157$).

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A STUDY OF THE EFFECTS OF PROTEOLYTIC ENZYMES AND $KBrO_3$ UPON THE VISCOSITY AND ALLIED PROPERTIES OF DISPERSIONS OF HARD RED SPRING WHEAT GLUTEN

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The use of viscosity determinations in the study of the effects of various factors upon protein characteristics is quite familiar to cereal technologists. The underlying principles of these methods rest presumably upon the concentration, configuration, and size of the protein particles.

Vogel and Bailey (1927) found the viscosities of leached and acidulated suspensions of durum wheat flour to be definitely lower than

like preparations of *vulgare* wheat flour of the same concentration. Morgan (1924) cited the utility of the viscosity test when applied to flour-water suspensions in determining flour quality.

Gortner (1924) developed a method whereby the viscosity of flour-water suspensions could be used to evaluate the quality of glutenin. Sharp and Gortner (1924) determined the viscosity of a number of acidulated dough-water suspensions made from fermenting dough. Samples of dough were taken at different periods during fermentation and the effect of fermentation upon gluten properties was determined. The viscosity of the suspensions following removal of electrolytes was found to increase as fermentation progressed. Malt flour and extract greatly diminished this effect owing to their proteolytic action.

Sharp and Gortner (1923) applied viscosity measurements of flour-water suspensions to the investigation of the comparative imbibitional properties of the flour protein. An effort was also made to correlate the colloidal properties of the protein with flour strength. These workers postulated that glutenin was the protein mainly responsible for the marked imbibition of gluten. An inherent difference in the physico-chemical properties of glutens from strong and weak flour was affirmed. These differences were found to be related to the colloidal state of the gluten proteins. Their report contains an excellent summary of literature dealing with viscosity measurements as applied to colloidal solutions. Blish and Sandstedt (1925) using more than 100 samples of Nebraska wheat flours studied the relationship between viscosity and baking quality. A relationship was found between actual viscosity and loaf volume. The order of this correlation was essentially the same as that between protein content and loaf volume. Actual viscosity and protein content were more highly correlated than other factors, and it was concluded that protein content forms as valuable a basis for the prediction of baking strength as the actual viscosity values.

Sharp (1926) investigated the plasticity of flour-water suspensions using a simple apparatus in which pressure from an ordinary hand-pressure pump was applied to force the material through a capillary. Various pressures were used. A particular flour-water suspension was found to be plastic when it contained 9% or more of flour by dry weight.

Johnson (1927) investigated the viscosity of flour-water suspensions at different extraction temperatures and various lengths of extraction period. The effect of degree of manipulation was also studied. The hydration value of flour particles was found to undergo a sharp change at 50° C. The rate of proteolytic activity of the flour suspension could be determined by viscosimetric techniques only in cases where

the degree of hydration and electrolyte content of the suspension were held constant when the viscosities of several suspensions digested for different periods of time were determined.

Johnson and Herrington (1928) found that the flour suspension which exhibited the highest viscosity contained approximately 50% of the total nitrogen. These workers also found (1928a) that certain proteins present in wheat flour caused a depression of the viscosity of acidulated flour suspensions.

Johnson, Herrington, and Scott (1929) measured proteolysis in digested flour-water suspensions using a method whereby the viscosity was determined after leaching and acidulating the suspension made from the residue following decantation of the supernatant liquid. The rate of decrease in viscosity was found to be greater at higher temperatures. The authors concluded that the formol titration method is probably the most convenient procedure for determining the proteoclastic activity of flour, but no information was yielded regarding hydrolytic changes of the protein micelle.

Johnson and Green (1930) showed the effect of fermentation upon the viscosity of dough-water suspensions. The rate of decrease in viscosity depended upon the H-ion concentration during fermentation, while the presence of NaCl and buffer salts prevented the decrease in viscosity during fermentation periods as long as 10 hours. This decrease in the absence of added salts was irreversible. Johnson (1931) proved that the viscosities of flour-water suspensions extracted with solutions of the potassium and sodium halides were higher than those of similar suspensions extracted with distilled water. The decrease in viscosity was according to the lyotropic series of the anions. Glutenin was not removed by the extraction with halides.

Jong and Klaar (1930) studied the viscosity changes induced in gliadin sols by varying alcohol and H-ion concentration. The viscosity of both positive and negative gliadin sols was found to increase up to 48% of alcohol by weight. Higher alcohol concentrations caused a marked decrease in viscosity for both positive and negative sols. The maximum viscosity was supposedly caused by the formation of an alcohol hydrate layer around the particles. Higher alcohol concentrations would remove this layer. The question whether an emulsoid will flocculate, unmix, or remain in solution by discharging apparently depends upon the isoelectric hydration.

Denham, Scott Blair, and Watts (1927) studied the utility of the Ostwald type of viscometer for purposes of measuring the viscosities of flour suspensions up to 30% concentration. This instrument proved to be entirely satisfactory provided it was calibrated with solutions of known viscosity, and the results were expressed in absolute units. It

was found that the capillary dimensions might be raised within wide limits without affecting the accuracy of the results, provided the flow is laminar within the range of the instrument, and that the critical velocity was not exceeded. Scott Blair, Watts, and Denham (1927) judged that viscosity measurements of flour suspensions at different concentrations would be useful for determining flour strength. An empirical formula ($\log \eta = k''c + d$) was found advantageous for plotting viscosity against concentration, k'' and d being constants. Plotting these values was found to yield a true straight line in all instances.

Bayfield (1933) reported the conclusions reached by the subcommittee on the viscosity tests for soft-wheat flour. This test, employing the MacMichael viscometer, is widely used by soft-wheat milling laboratories and is generally considered to be of distinct value in connection with quality evaluation. By means of this test it appeared probable that relatively small differences due to variety of wheat, ash, and protein content might be detected. Later Bayfield (1935) postulated that protein quantity was readily measured by viscosity determinations. Quality of protein, however, proved to be more difficult to determine, although when several wheat varieties were compared marked differences were observed.

Rose and Cook (1935) found that the viscosity as determined on protein dispersions with the U-tube viscometer yielded valuable information regarding particle size, hydration and denaturation changes in these dispersions when they were prepared by different methods.

Landis and Frey (1938) described a method for measuring proteolytic activity through changes in the rate of gelation of gelatin sols following exposure to proteolytic agents. Harris (1938) published the conclusions derived from a study of the effects of various proteoclastic enzymes upon wheat-gluten "solubility" in sodium salicylate solution. It was shown that in the majority of experiments enzyme action markedly hastened the dispersion of the gluten, and this effect increased with an increase in concentration of enzyme. Inhibition of enzymic activity by $KBrO_3$ was demonstrated, especially in the instance of papain and bromelin. Later this study was extended to investigations into the effects upon the relative distribution of the protein fraction removed from sodium salicylate gluten dispersions by additions of $MgSO_4$. Striking changes in the proportion of protein removed in the first fraction were evident when papain, malt, and taka-diastase were added to the dough mix from which the glutens were subsequently washed. This effect was not evident in the pepsin treatment. Balls and Hale (1938) applied a viscosity method with gelatin to the measurement of the proteolytic activity of wheat-flour proteinase prepared by them.

In view of the substantial amount of work which has been done with flour-water suspensions in an endeavor to relate viscosity values to flour quality, it was judged expedient to apply such methods to the elucidation of viscosity changes in connection with the action of proteolytic enzymes upon gluten washed from doughs treated with such enzymes. The Ostwald pipette is a convenient form of capillary instrument to use for this purpose. Relatively small quantities of liquid are required for a reading, the pipette and contents are easily held at a constant temperature, and readings covering a substantial range in viscosity may be made with one instrument. Mangels and Bailey (1933) and Mangels (1934, 1936) used the Ostwald pipette for purposes of studying the viscosities of a large number of starches prepared from hard red spring and durum wheats with satisfactory results.

Experimental Material and Methods

A commercially milled hard red spring wheat flour was used in this investigation. The flour had been neither bleached nor diastated in the mill and contained 13.1% protein and 0.51% ash on a 13.5% moisture basis. The malt-phosphate-bromate loaf volume was 625 cc.

The glutens were washed from doughs mixed in the usual manner for baking, using a two-minute mix in the Hobart-Swanson mixer. The formula used included 1% salt, 3% yeast, 5% sucrose, and distilled water to firm consistency. The glutens were washed immediately after mixing, under a small standardized stream of 0.1% sodium phosphate which had a pH of 6.8. This procedure corresponds essentially to the technique formerly used by the senior author in work of this nature and already reported (Harris, 1938). The gluten was divided and placed in 10% sodium salicylate solution with thorough shaking. The concentration used in the first part of this work was 6 g. of wet crude gluten per 100 cc. of sodium salicylate solution. Shaking was continued at 5-minute intervals and aliquots taken for viscosity determinations at hourly intervals for the first two hours, then at two-hour intervals until the course of dispersion had been clearly established. Approximately 35 cc. of liquid were decanted into 50-cc. centrifuge tubes from this suspension following a settling period of five minutes from the last shaking to allow suspended shreds and particles of gluten to settle. These aliquots were then centrifuged and the density and viscosity determined. After these determinations the liquid as well as the residual material deposited in the centrifuge tubes was returned to the original dispersion flask. In this way no permanent change in the concentration of the liquid in relation to the undispersed material was caused by removal

of the aliquot. Since only 5 cc. of liquid were required for the measurement of viscosity, the bulk of the colloidal solution was returned when the density had been ascertained. By this technique, the total 35 cc. were separated from the actual mixture only during the centrifuging and the weighing of the specific gravity bottle.

The viscosity determinations were carried out with an Ostwald pipette. This pipette, and the dispersions preliminary to testing, were held at $25 \pm 0.1°$ C. in a constant-temperature bath fitted with heater, thermoelectric regulator, and stirring device. This apparatus

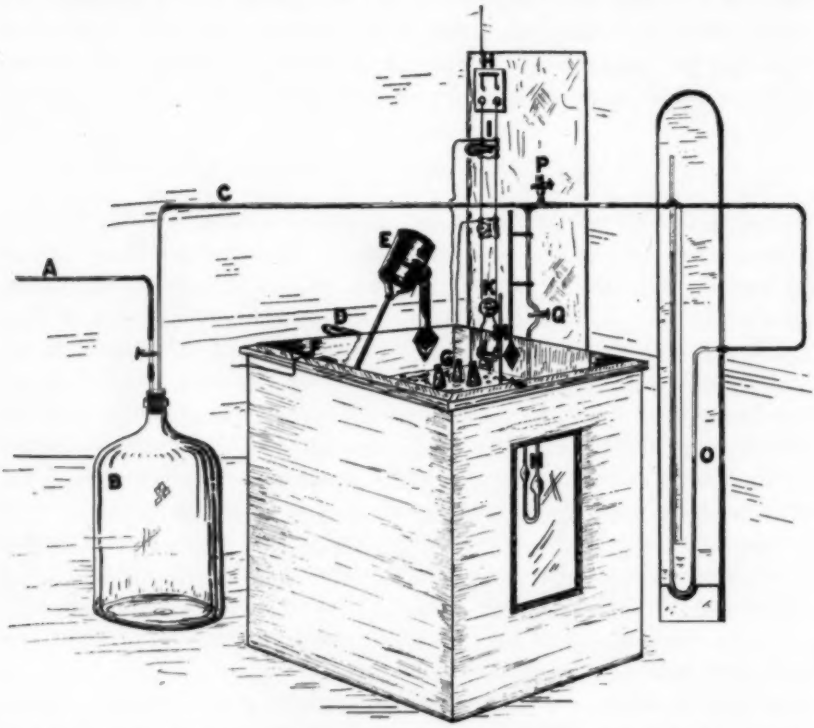

Fig. 1. Illustration of constant-temperature bath and accessory apparatus used in determining the viscosities and rates of flow.

is shown in Figure 1. The bath itself was constructed of wood and lined with sheet copper. Glass windows were installed at each end of the bath. Above the window at the left in the illustration a light and reflector (*D*) served to illuminate the Ostwald pipette (*N*) which could then be read through the window shown on the right in the figure. A copper shelf (*G*) served both as baffle plate and as a holder for flasks containing the various liquids and dispersions worked with, to insure that the contents were held at the temperature of the bath. The bath and contents were kept at a constant temperature by a

thermoregulator (*F*) which was placed in parallel with the pilot light (*I*), heater switch (*K*), and heater (*L*). A switch (*H*) led to the source of current. The water was kept in circulation by a stirrer (*E*). The difference between atmospheric pressure and a small partial vacuum was used as a driving head to force the liquid through the pipette rather than the application of direct pressure upon the liquid. It is assumed that the partial vacuum which served to draw the liquid through the capillary of the Ostwald pipette was equivalent to an applied pressure which would force the liquid through. To accomplish this purpose, a large carboy (*B*) was partially evacuated by a glass tube (*A*) which led to a water-suction pump. A glass tube (*C*) united the carboy to the mercury manometer (*O*) and to the pipette through a four-way glass tube and suitable connections. The degree of vacuum obtained was read from the manometer. A valve (*P*) served to release the vacuum in the system when the determination was finished. The three-way valve (*Q*) was used to connect the pipette to the partial vacuum and to the air.

A series of pressures was used on each liquid examined. The mode of procedure was to start at a low partial vacuum and raise the pressure by small increments between successive determinations. The pressure used varied from the weight of the liquid in the bulb of the pipette to the same weight increased by approximately 100 mm. of mercury. When measuring the increase in viscosity with time of dispersion, the pipette was disconnected from the glass tube and two-way valve, thus insuring free flow of liquid without extraneous resistance in the pipette capillary.

Discussion

The effects of different papain concentrations in comparison with 0.004% and 0.016% increments of $KBrO_3$ upon the viscosities of the gluten dispersions are shown in Table I. These determinations were

TABLE I

EFFECT OF VARIOUS ADDITIONS OF PAPAIN AND POTASSIUM BROMATE TO DOUGHS UPON VISCOSITY CHANGES IN GLUTEN DISPERSIONS

Dough treatment	Viscosity (centipoises $\times 10^2$)								
	1st hr.	2nd hr.	4th hr.	6th hr.	8th hr.	10th hr.	12th hr.	Final	dc/dt[1]
Control	131.1	137.9	155.2	172.8	182.9	190.3	197.4	219.9	8.5
Papain 0.004%	132.0	139.2	161.1	184.7	195.3	199.1	203.3	203.9	9.6
Papain 0.004% + $KBrO_3$ 0.004%	128.6	135.7	158.9	179.2	193.2	201.8	209.5	192.4	8.5
Papain 0.004% + $KBrO_3$ 0.016%	127.8	131.5	149.9	168.5	186.2	197.2	206.5	197.6	7.0
Papain 0.008%	135.3	150.7	182.6	197.7	197.3	194.8	194.8	188.2	13.4
Papain 0.008% + $KBrO_3$ 0.004%	129.5	140.6	168.0	188.9	198.8	198.6	198.3	188.9	11.9
Papain 0.008% + $KBrO_3$ 0.016%	129.2	134.3	160.8	181.8	194.8	197.7	201.1	184.8	9.2
Papain 0.02%	168.7	170.1	169.8	170.7	170.6	169.0	169.0	166.6	41.4
Papain 0.02% + $KBrO_3$ 0.004%	162.1	171.7	167.1	167.0	167.0	167.0	167.0	161.3	34.8
Papain 0.02% + $KBrO_3$ 0.016%	152.6	174.6	169.4	168.5	168.5	168.5	168.1	163.5	25.3
Papain 0.03%	162.0	161.0	160.0	161.2	158.9	159.6	159.6	160.5	

[1] Calculated by use of interpolated values from Figure 2.

run over a period of 12 hours. The results are also shown graphically in Figure 2, where viscosity is plotted against time. Owing to the nature of the data, the results are more easily interpreted from the figure and the discussion will be largely confined to it, rather than the table. From the results obtained with papain alone it will be noted that the effect of the enzyme is markedly to increase the viscosity during the initial hours of dispersion. This increase becomes more

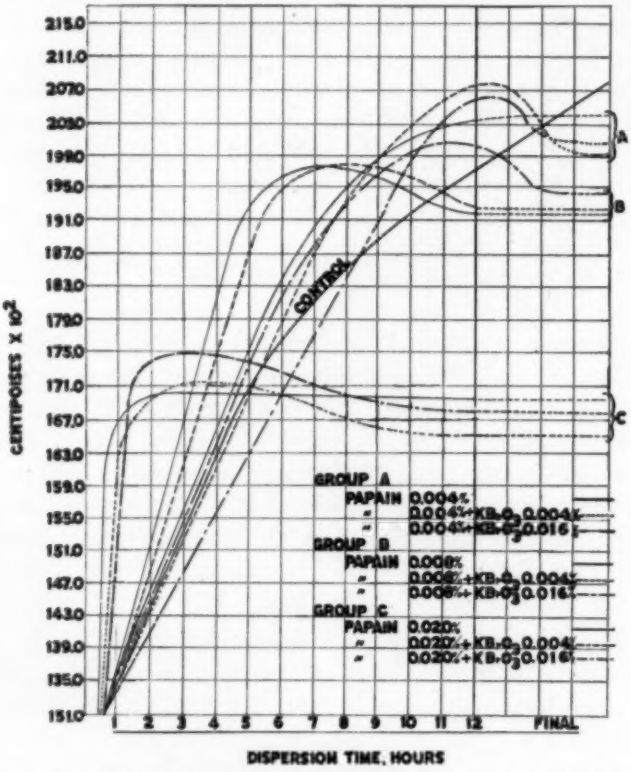

Fig. 2. Effects of papain and potassium bromate upon the increase of viscosity with time of dispersion in gluten dispersions.

marked at higher papain dosages. The effect reaches a maximum and tends, in some instances, to decrease slightly at the final hour, although this decrease is probably not significant. When $KBrO_3$ was also added to the doughs, in addition to papain, a lower rate of dispersion was at first evident in every instance. These results of bromate action upon papain are also shown in Table I under the column headed dc/dt, which represents rate of change with increase in time. It is apparent from these results that papain by itself greatly increases this value, while bromate in each instance lowers the value obtained in the unbromated

papain treatment. The fraction dc/dt represents the slope of the lines from the starting point until the time when the line begins to curve, showing the effect of other factors such as structure, configuration, and size of particle upon the viscosity, rather than increase in gluten concentration in the dispersion. This fraction was calculated with interpolated values taken from the figures. The equation representing the slope, or the rate of disintegration, is:

$$\frac{dc}{dt} = \frac{\eta a - \eta o}{ta - to},$$

where: ηa = viscosity at the point where the line begins to curve.
ηo = viscosity at the zero hour—27.3.
ta = time corresponding to ηa.
to = time corresponding to ηo or 0.

In the majority of cases, the final viscosity of the bromated dough-gluten dispersions fell below the unbromated dough-gluten value and would tend to indicate some effect upon the particle size or properties, corresponding to a slight activation of the papain. This is especially noticeable in the lowest 0.004% bromate treatment, but is not apparent in the intermediate 0.008% papain-treated dough gluten. This effect of $KBrO_3$ upon viscosity of papain-treated doughs will be discussed in more detail a little later in this paper.

In Table II similar data obtained with three increments of pepsin and corresponding bromate dosages are shown, while Figure 3 presents

TABLE II

EFFECT OF VARIOUS ADDITIONS OF PEPSIN AND POTASSIUM BROMATE TO DOUGHS UPON VISCOSITY CHANGES IN GLUTEN DISPERSIONS

Dough treatment	Viscosity (centipoises × 10^3)								
	1st hr.	2nd hr.	4th hr.	6th hr.	8th hr.	10th hr.	12th hr.	Final	dc/dt[1]
Control	131.1	137.8	158.1	189.1	216.5	224.3	231.0	233.4	9.8
Pepsin 0.1%	132.2	135.2	154.7	178.0	200.9	213.7	223.0	238.7	8.8
Pepsin 0.1% + $KBrO_3$ 0.004%	134.8	136.5	159.0	177.4	201.6	215.1	220.1	232.7	9.1
Pepsin 0.1% + $KBrO_3$ 0.016%	132.1	136.3	148.9	165.8	190.6	200.1	211.8	233.1	7.9
Pepsin 0.3%	139.2	158.8	195.2	210.4	218.1	219.6	219.3	225.6	16.9
Pepsin 0.3% + $KBrO_3$ 0.004%	149.9	179.5	206.3	215.1	216.5	220.9	220.9	210.4	21.9
Pepsin 0.3% + $KBrO_3$ 0.016%	140.7	159.5	189.8	207.1	215.5	222.7	222.7	220.9	14.9
Pepsin 0.5%	194.3	197.4	196.4	197.0	199.0	199.0	199.0	196.5	67.0
Pepsin 0.5% + $KBrO_3$ 0.004%	197.1	201.9	199.8	199.9	199.9	199.9	199.9	199.7	69.8
Pepsin 0.5% + $KBrO_3$ 0.016%	191.5	201.0	199.6	199.7	199.7	199.7	199.7	200.4	64.4

[1] Calculated by use of interpolated values from Figure 3.

the same data in graphic form. Here the increase in rate of dispersion with increase in enzyme concentration is similar to that shown in the papain data. That is, increase in enzyme treatment causes an increase in rate of dispersion with one exception: in the instance of 0.1% pepsin

dosage, the dispersion of the gluten appears to be retarded, pointing, it would appear, to a coagulating effect of this concentration of the enzyme upon the flour gluten. A general conception of enzyme action

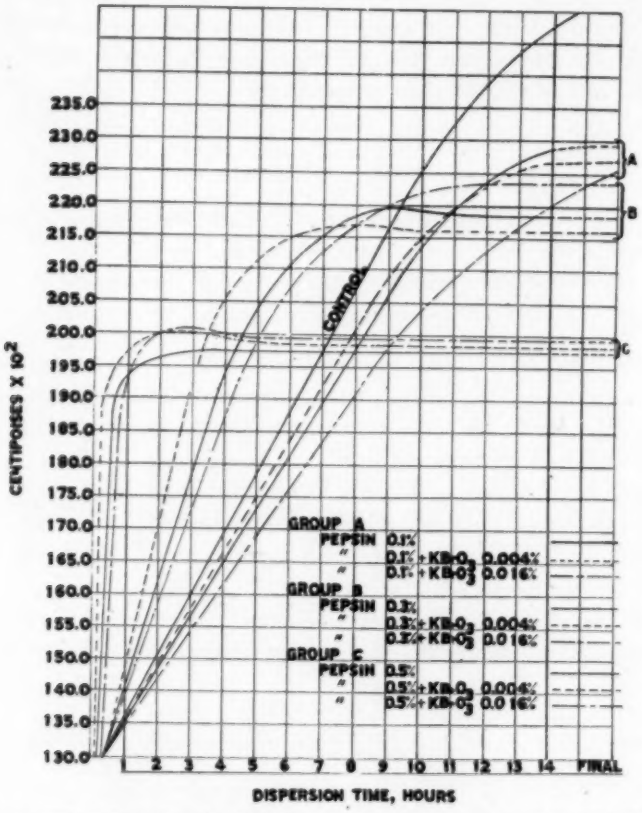

Fig. 3. Effect of pepsin and potassium bromate upon the increase of viscosity with time of dispersion in gluten dispersions.

is that a preliminary coagulation of protein is followed later by disintegration and dispersion. This effect was not noticeable in the case of papain treatments without bromate probably owing to the more drastic action of the latter enzyme upon flour gluten. When we consider the effect of bromate upon pepsin, certain deviations from its effect upon papain are noted. In the case of pepsin, a distinct increase in rate of dispersion is shown for the three concentrations of 0.004% $KBrO_3$, while the heavier concentrations of 0.016% $KBrO_3$ repress or inhibit the rate of dispersion. These effects, in the writers' opinion, are due first to an activating effect upon the enzyme exerted by the lower bromate concentration and second to an inhibitory effect upon the enzyme of the higher 0.016% bromate treatment. When com-

paring the results of bromate upon these two enzymes, one must remember that papain is more drastic in its action upon flour gluten than pepsin and the stimulating effects of lower dosages of bromate are not of sufficient magnitude to be revealed in the data dealing with rate of dispersion with the former enzyme. These conclusions are verified mathematically by the values for dc/dt, which show an initial increase with bromate as compared with the unbromated enzyme treatment followed by a decrease in each instance at the higher bromate concentration. These differences in action of $KBrO_3$ upon these two enzymes as well as the apparent coagulating effect of low dosages of pepsin are in line with former conclusions reached by the senior author.

Falk and Winslow (1918) also found indications of the possibility of potassium bromate exerting a stimulative action upon the proteolytic enzymes during dough fermentation. These workers found that a concentration of one part bromate in 100,000 to one part in 200,000 appeared to stimulate the digestion of casein by trypsin or pancreatin but when the bromate concentration was increased to one part in 10,000 a slight inhibition of enzymic activity was noted.

The viscosities were calculated from an adaptation of the law of Poiseuille:

$$\eta = \frac{[(d_1)(h) + (\Delta p)d_2]t_2}{hd_3t_1},$$

where d_1 equals density of the liquid as determined by a standardized picnometer, h equals height of liquid in the viscosimeter, Δp is pressure applied, expressed in cms. of mercury, d_2 is the density of mercury (13.6), t_2 is time of flow at corresponding pressure, d_3 is density of the reference liquid, and t_1 is the time in seconds of the reference liquid. These gluten dispersions were all adjusted to a crude protein content of 2500 mg. per 100 cc. of dispersion. The data obtained on the various liquids using pressure differences in determining the viscosity were calculated as rate of flow ($1/t \times 10^2$), t being expressed in seconds; viscosity (in centipoises) $\times 10^2$; and viscosity/$t \times 10^2$. The latter quantity was computed as a test of the experimental precision of the viscosity tests. In the construction of Figures 4 and 5 the entire set of results obtained was used to obtain as true a picture of the relationships involved as possible.

The data derived from the $KBrO_3$ treatments with application of pressure were not depicted graphically, because of difficulties in presenting the slight differences obtained between the bromated and unbromated doughs. Differences of little significance were found between the bromated and the unbromated dough gluten dispersions,

although in the instances of 0.004% papain the lower concentration of bromate appeared to increase the rate of flow.

The data relating to rate of flow, as influenced by pressure and presented in Figure 4, show that water has the highest increase in rate of flow with increase in pressure. From the papain data it is apparent that papain significantly increased the rate of flow of the dispersed

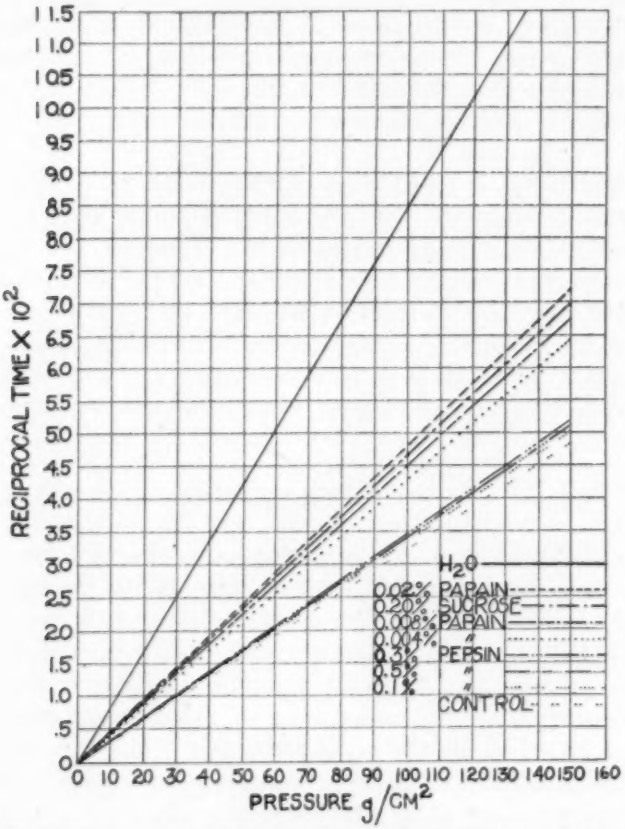

Fig. 4. Effects of pressure upon rate of flow in various liquids and dough gluten dispersions. Data show no evidence of plastic flow. Curves apparently all pass through point of origin.

gluten prepared from doughs to which this enzyme was added. This effect increased with increasing concentration of enzyme. It is also apparent that pepsin increased the rate of flow of dough gluten dispersions in contrast to the untreated dough dispersions, but this effect was much less than in the case of papain and points to fundamental differences in the mode of attack of these enzymes upon flour gluten. The differences shown for the various concentrations for pepsin are scarcely significant. As these curves all pass through the point of

origin, it is evident that plastic flow does not exist and we have true viscous flow. The data depicted in Figure 5 show quite clearly the effect of the two proteolytic enzymes in lowering the viscosity with pressure when the dispersion of the gluten has apparently reached a constant or final value. It will be noticed that the untreated control dough has the highest viscosity curve, followed in order of decreasing viscosity by 0.1%, 0.3%, and 0.5% pepsin respectively.

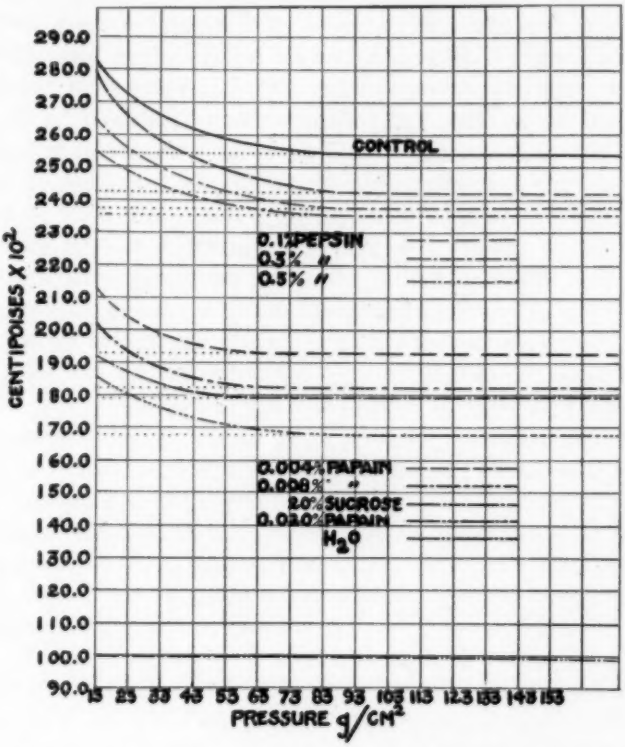

Fig. 5. Effect of pressure upon viscosity of the various gluten protein dispersions including 20% sucrose and redistilled H_2O for comparison.

The papain curves all show markedly lower viscosities than the curves already mentioned and decrease in order of increasing papain treatment. These results apparently indicate that the structure of the protein micelle has been substantially altered by these two enzymes, especially by papain. It is extremely probable that the size of the protein micelle has been reduced. This particle size reduction appears to be particularly marked in the case of papain and accordingly this enzyme must cleave the gluten complex into smaller portions, whereas the action of pepsin does not progress to nearly the same

degree and particles formed in the instance of this enzyme are much larger in size. It is accordingly probable that the fundamental bonds uniting the various portions of the gluten complex have not been attacked to any extent.

From the viscosity pressure relationships shown in Figure 4, it is evident that water is the only liquid which gives a linear relationship, the other curves being distinctly curvilinear over the initial pressure range. From the equation of Poiseuille ($\eta = \pi p R^4 t/8lv$) it follows that a decrease in the effective radius R of the capillary will cause a decrease in viscosity. If a decrease of a very small magnitude were effected through the agency of the initial increment of pressure, the result would be an apparent fall in viscosity followed by a constant value for the relationship of this variable with pressure. The final outcome of such a phenomenon would be a curve showing an initial inflection, then shading off into a straight line. It is difficult to picture such a situation arising, however, in the instance of 20% sucrose solution. It was also found that viscosity measured with capillaries of different bores gave the same type of curve when viscosity was plotted against pressure, and this evidence would appear to be against the theory of effective radius change, inasmuch as the magnitude of the change would vary for different capillary radii.

A more plausible explanation is by means of the following considerations. The size of the meniscus of the liquid is changed as the liquid leaves the upper capillary and enters the bulb of the Ostwald pipette. Turbulent flow probably occurs at first with application of pressure to the system, to be later replaced by laminar flow as the pressure is further increased. Resistance is also likely to occur at the lower end of the capillary, which is submerged in the liquid contained in the lower portion of the pipette. The sum total of these effects would be to cause a preliminary loss of energy from the system. This effect would correspond to a decrease in pressure, with apparent decrease of viscosity. As the pressure is further increased these effects are overcome and a linear relationship then results between viscosity and pressure. Liquids with relatively low surface tension, such as water, will not show this effect.

Part of the data were recalculated in terms of viscosity/time and these values are shown in Figure 6. A linear relationship should represent the relationships between the variables shown. A departure from linearity, or a scattering of the dots, would indicate a degree of experimental error or lack of precision in the work. It will be noted from a careful inspection of this figure that the dots do fall very closely on a straight line and that the projection of this line would pass very nearly through the origin. One or two of the variables, especially with

the 20% sucrose solution, do show some displacement from the straight line, which passes through the majority of the points. In a number of cases these dots fell together, and it was therefore necessary to displace several of the points somewhat in order to place them on the figure. This of course would cause a slight distortion in the general appearance of the curve.

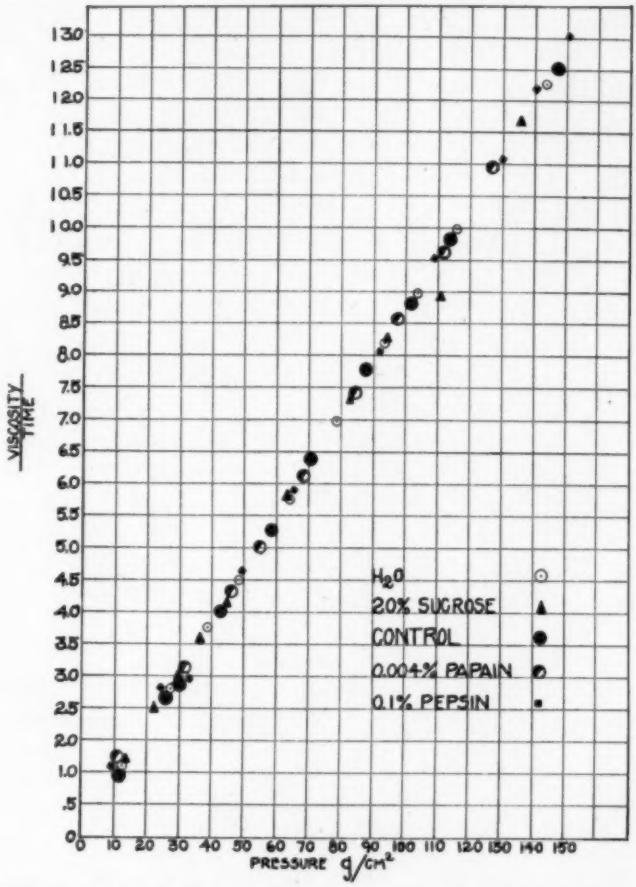

Fig. 6. Relation of the ratio, viscosity/time of flow, to pressure. This is a test of experimental precision and in this instance shows little evidence of any substantial error in viscosity determinations.

Table III contains the data obtained by fractionating the gluten protein from the dispersions following the determination of the relationships between pressure, rate of flow, and viscosity. The effect of increasing the papain dosage is shown in a marked fall in the quantity of protein removed from the dispersion, and corresponds with former results published by the senior author. Pepsin, on the other hand, shows little effect upon the protein fractionated. These results are

TABLE III

EFFECT OF INCREMENTS OF $KBrO_3$ UPON THE QUANTITY OF PROTEIN FRACTIONATED FROM 10% SODIUM SALICYLATE DISPERSIONS OF GLUTEN WASHED FROM DOUGHS MADE UP WITH ADDITIONS OF PAPAIN AND PEPSIN [1]

Results expressed in mg. protein per 100 cc. dispersion [2]

Treatment	$KBrO_3$ %		
	0.0	0.004	0.016
Control (No enzyme added)	1630	—	—
Papain 0.004%	997	986	835
Papain 0.008%	648	658	638
Papain 0.02%	392	319	361
Pepsin 0.1%	1590	1602	1642
Pepsin 0.3%	1613	1545	1607
Pepsin 0.5%	1596	1585	1516

[1] Results show quantity of protein precipitated by 6 cc. of concentrated $MgSO_4$ solution.
[2] Dispersions adjusted to a protein concentration of 2500 mg. per 100 cc.

depicted graphically in Figure 7. These papain relationships are similar to former data published by Harris (1938) using lower gluten protein concentrations (not on a definite protein basis) in the dispersion. These fractionation results correspond roughly to the final viscosities of these dispersions and offer further evidence of the comparative effects of papain and pepsin upon particle size in these dispersions. As papain concentrations increase, particle size decreases and less protein is removed by the fractionation procedure. In the case of pepsin, little effect upon particle size occurs and accordingly little change is evident in the fractionation value.

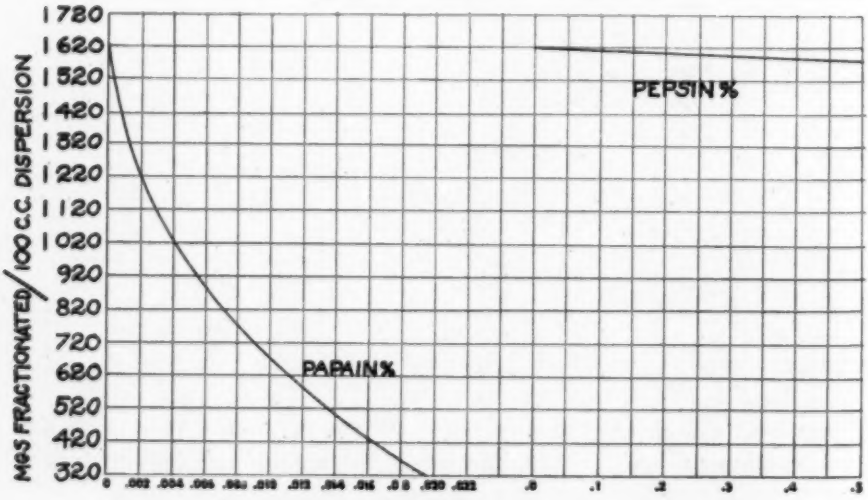

Fig. 7. Comparative effects of increments of papain and pepsin upon the quantity of protein fractionated from dough gluten dispersions.

The effect of $KBrO_3$ upon fractionation is also shown in Table III and is represented graphically in Figure 8. In discussing first the effect of bromate upon papain, it appears that the lowest concentration of enzyme, namely 0.004%, is activated by bromate as the quantity of protein removed is diminished by the bromate, which corresponds

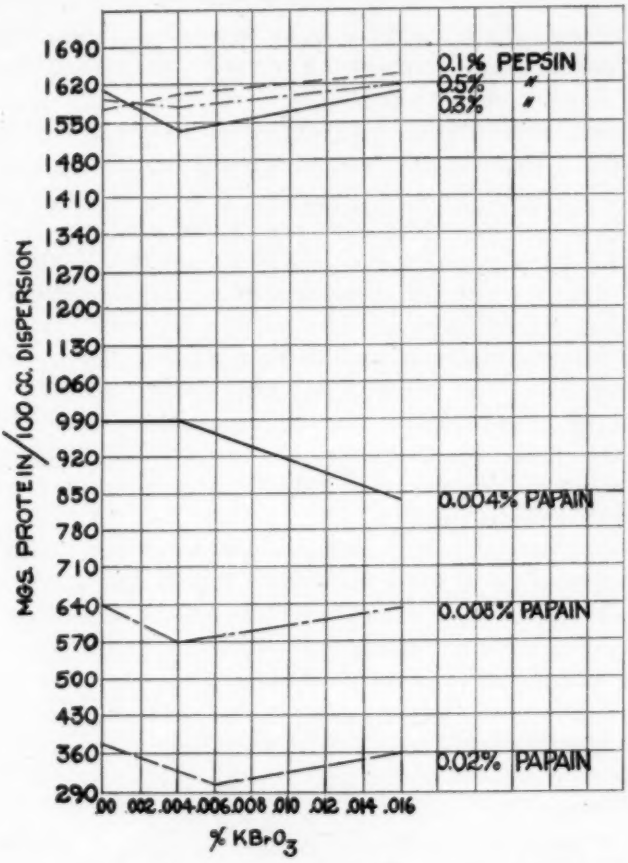

Fig. 8. Comparative effects of increments of potassium bromate upon various concentrations of papain and pepsin flour doughs as registered in mgs. of protein fractionated from dough gluten dispersions.

to the effect of increasing the enzyme concentration, as shown in Figure 7. When the papain concentration is increased, however, the lower dosage of bromate activates, while the heavier dosage represses. Bromate appears to have little effect upon pepsin, and this result would be expected with pepsin, considering that this enzyme does not appear to change the particle size materially in these dispersions.

Summary and Conclusions

Glutens were washed from a large number of doughs prepared from a hard red spring wheat flour. Various quantities of papain and pepsin were added to these doughs. Potassium bromate was also used in addition to the enzymes in increments of 0.004% and 0.016%. Control doughs were run for comparative purposes. The glutens were washed immediately after the doughs were mixed and dispersed in 10% sodium salicylate solution. The rate of dispersion was determined by viscosity measurements.

The results obtained indicated a marked rise in viscosity with time of dispersion. This rise was distinctly greater in most instances in the dispersions prepared from doughs treated with enzymes, although there was some evidence that a low concentration of pepsin decreased the rate of dispersion, probably owing to a slight coagulation of gluten protein. Bromate retarded dispersion in the papain-treated doughs, but increased the rate for pepsin when present in 0.004% concentration. A further investigation was conducted, using a higher concentration of dispersed gluten and adjusting the final concentration to 2500 mg. of protein per 100 cc. Viscosities were then run on these dispersions with suitable pressure variations. The quantity of protein fractionated by adding 0.6 cc. of concentrated $MgSO_4$ solution to 10 cc. of the colloidal solution was then found.

The data obtained by the method showed that apparently viscous flow was present in all the dispersions. Papain had a greater effect than pepsin in increasing the rate of flow and in decreasing the viscosity, pointing it would appear to the production of smaller particles caused by differences in protein cleavage by the two enzymes. Bromate did not show any marked effect in this instance. The fractionation data also gave evidence of papain activity in reducing particle size with increasing enzyme concentration, while pepsin had little effect. Some evidence of a slight activation of papain at 0.004% concentration by bromate was noted, followed by repression as the papain concentration increased.

Acknowledgment

The authors wish to express their sincere appreciation of valuable advice contributed by Dr. R. A. Gortner and Dr. Dallas Dedrick regarding the viscosity determinations. Acknowledgment is also made of the aid of John Monge in working up the data and constructing the graphs and of technical assistance from WPA and NYA funds.

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A COMPARATIVE STUDY OF SOME PROPERTIES OF DRIED GLUTENS PREPARED FROM VARIOUS TYPES OF WHEAT

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(Received for publication July 13, 1939)

Several years ago the writer, in collaboration with H. N. Bergsteinsson,[1] built up the protein content of a soft-wheat flour dough by additions of freshly washed wheat gluten prepared from hard red spring wheat flour. The gluten was reduced to small shreds by hand and added to the flour and other dough ingredients in the mixing bowl. It was then incorporated into the dough by careful mixing. The moisture and protein content of the gluten were determined and from these data the final protein level of the resultant blend could be calculated. The levels used were 12.1%, 16.7%, and 21.3%, built up upon a 7.5% protein flour. These values are on a 13.5% moisture basis. The baking formula used was the malt-phosphate-bromate, superimposed upon the A.A.C.C. standard basic. Photographs of the resultant four loaves are shown in Figures 1 and 2, which depict the size, external appearance, and interior characteristics of these loaves. It is quite evident that marked improvements in these loaf attributes resulted from the addition of wet crude gluten to the soft-wheat flour. It is very probable that further differentiation, among the loaves, especially in volume, would have been secured if larger increments of $KBrO_3$ had been used for the higher protein levels.

Aitken and Geddes (1938) devised a method whereby the protein contents of flours could be adjusted to any desired level through the addition of dried and ground gluten, thereby obviating possible difficulties in properly incorporating wet crude gluten in the dough and facilitating the determination of the quantity required for any desired

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protein level. In addition, the danger of changes in wheat gluten properties during the time required for moisture and protein determinations was eliminated. These workers used a low drying temperature of 32°C. and a rapid air flow through a falling humidity gradient. The gluten was then reduced to flour-like fineness before using. The addition of such gluten flour to a series of weak, intermediate, and strong world wheats to equalize their protein contents resulted in a marked improvement in flour strength but the crumb color was impaired. From their results the authors concluded that the majority of world wheats possessed glutens of similar character, and their varying

Fig. 1. External appearance of loaves baked from blends of hard red spring wheat gluten and soft wheat flour. Gluten used in original undried form.

Fig. 2. Internal appearance of loaves baked from blends of hard red spring wheat gluten and soft wheat flour. Gluten used in original undried form.

strength characteristics were due to protein-content differences. Some wheats of English, German, and Italian origin are deficient in baking quality owing to lack of satisfactory protein content, combined with a weak and unsatisfactory nature of the gluten. It was noticed that the degree of dispersion in 0.05*N* acetic acid had been somewhat lowered as a result of drying. In view of the results obtained by Aitken and Geddes with dried gluten, the author decided to investigate the effects of additions of gluten flour prepared from different classes and varieties of American wheats and prepared, as far as feasible, by their method. It was decided to determine the effects, as far as possible, of the drying

process upon the properties of these glutens using gluten dispersions in 10% sodium salicylate solution as described by me in former publications (1938, 1939), and by Harris and Johnson (1939, 1940).

Material and Methods

Fourteen lots of flour containing both commercially and experimentally milled samples from different types and varieties of wheat were included in this investigation. This series covered as wide a range in baking strength and genetic variability as it was possible to secure, and should, therefore, yield pertinent information regarding the relationships of gluten properties to these two variables.

Durum wheats were also included, because of their commercial importance in North Dakota, with the aim of obtaining information upon the gluten characteristics of these wheats. A soft-wheat flour commercially milled from Pacific Coast wheat containing 7.5% protein was used as a basic flour upon which the various gluten flours were superimposed. The malt-phosphate-bromate formula [2] was used throughout the baking test. The original flours were also baked by this formula.

In preparing the glutens, the flours were mixed into a stiff dough with 0.1% sodium phosphate solution (pH 6.8) in the Hobart-Swanson, and allowed to stand a short time at room temperature under tap water. The glutens were washed under a small stream of sodium phosphate solution, covered with the same solution for approximately 15 to 30 minutes, then divided into two- to three-gram portions and placed upon sheets of waxed paper for drying. The sheets of gluten were placed in a fermentation cabinet equipped with a temperature control, a Bahnson humidifier, and an electric fan to insure thorough air circulation. The drying temperature was held at 32 ± 2°C. and the humidity gradient was gradually lowered manually.

At the end of 24 hours, the gluten pieces appeared to be sufficiently dried and were removed from the waxed paper and reduced to flour in the micro mill. Moisture, ash, and protein determinations were run on these glutens. The quantity of dried gluten required to raise the protein level of the soft-wheat flour to 14.0% (13.5% moisture level) was calculated in each case. The bakings were made in triplicate and the results averaged. The doughs were mixed for three minutes in the Hobart-Swanson with sufficient water to produce normal dough consistency. The dried gluten appeared to be entirely incorporated in the dough and no streaks or dark spots were visible in the baked loaves. In addition samples of wet crude gluten, the dried gluten

[2] This formula contained 0.3% diastatic malt, 0.1% ammonium phosphate, and 0.001% $KBrO_3$ superimposed upon the A.A.C.C. basic formula which contained 5% sucrose instead of the customary 2.5%.

powder, and gluten reformed from the dried gluten by suitable additions of sodium phosphate solution were dispersed in sodium salicylate, the protein content of the dispersions was adjusted to a concentration of 1400 mg. per 100 cc., and the viscosity [2] and protein fraction were determined. In this way a comparison was obtained between the three gluten dispersions in regard to these properties, as well as with respect to apparent solubility in the dispersing agent, visual appearances of the dispersion, etc., in order to permit conclusions regarding any changes induced in the washed gluten by the drying method.

Discussion

A description of the flours used in this investigation, with ash, crude protein, and baking data, is presented in Table I. The results

TABLE I

PROTEIN, ASH AND BAKING DATA ON FLOURS USED IN THIS INVESTIGATION [1]
(Arranged in order of increasing flour protein)

Sample No.	Type of flour	Ash	Crude protein (N×5.7)	Loaf volume	Color	Texture
		%	%	cc.	%	%
1	Pacific Coast soft	0.32	7.5	359	96.0	89.3
2	Soft red winter	0.48	8.5	469	92.5	94.0
3	Hard red winter (Chiefkan)	0.48	10.5	525	92.5	93.5
4	Yaroslav emmer	0.58	12.2	440	91.0	93.0
5	Hard red winter	0.51	12.3	616	93.5	92.5
6	Hard red spring (Nordhaugen)	0.40	12.6	606	94.5	92.0
7	Durum (Monad)	0.72	12.7	525	94.5	93.0
8	Durum (Mindum X Vernel)	0.66	13.0	417	91.0	93.0
9	Hard red spring (commercially milled)	0.51	13.1	695	94.5	93.0
10	Durum (Pentad)	0.69	13.4	465	90.0	93.0
11	Hard red spring (Thatcher)	0.37	13.5	695	94.5	92.5
12	B & S duster bottom, mill-stream	1.74	16.6	570	89.0	90.0
13	3rd break, mill-stream (commercially milled)	0.66	17.0	692	92.0	91.0
14	Durum (Mindum)	0.88	17.1	465	90.0	93.0
15	5th break, mill-stream (commercially milled)	1.35	19.4	645	89.0	90.0
Average			13.7	559	92.0	92.4

[1] On 13.5% moisture basis.

are arranged in order of increasing protein content, inasmuch as this factor is an important index of flour strength. It is evident from this table that a wide range of strength existed among these flours. The four principal wheat types grown in the continental United States are included, as well as several commercial mill-stream flours of relatively high protein content. A number of durums and an emmer were

[2] Viscosities were determined by an Ostwald pipette at 25 C. Densities were measured with a picnometer standardized against redistilled water.

included in the experimental material to furnish information in respect to the gluten properties of these wheats and their relationships to baking strength. The protein content ranged from 8.5% to 19.4%, a difference of 10.9%, while the loaf volume varied from 417 cc. to 695 cc. Flour No. 1, which had the lowest protein content and loaf volume, was used as a basic flour to which the dried gluten was added. Several of the durum wheat flours gave loaves of inferior volume, as would be expected, and substantially lowered the average loaf volume for this set of flours. These durum results also tended to lower the relationship between protein content and loaf volume. A large variability in flour ash existed among the various samples of flour.

In Table II are shown the comparative data obtained from the blends of soft-wheat flour and dried gluten. More variability was

TABLE II

PROTEIN CONTENT AND BAKING DATA ON BLENDS OF SOFT-WHEAT FLOUR AND DRIED GLUTEN

No. of flour from which gluten was prepared	Crude protein (N×5.7)	Loaf volume	Color	Texture	Variation of loaf vol. from original
	%	cc.	%	%	cc.
1 Control, no gluten added	7.5	359	96.0	89.3	—
2	13.7	467	95.0	95.0	+ 2
3	13.5	513	93.0	94.0	+ 12
4	13.8	488	92.0	94.0	− 48
5	13.9	494	95.3	96.3	+122
6	14.0	481	95.3	94.3	+125
7	13.3	476	94.0	94.0	+ 49
8	13.5	492	92.0	93.1	+ 75
9	14.0	469	95.3	93.7	+226
10	14.0	482	93.0	94.0	− 17
11	14.1	453	95.0	95.0	+242
12	13.7	397	92.0	94.0	+173
13	13.5	456	94.0	95.0	+236
14	14.0	474	94.3	94.7	− 9
15	13.5	482	94.0	94.0	+163
Average of blends	13.8	473	93.9	94.4	+ 96.5

evident in the protein contents of these blends then was desired, but it is probable that this had little effect upon the final results obtained. The loaf volumes did not differ greatly from the general level except in the instance of Chiefkan and the B and S duster bottom flour blends. Why Chiefkan gluten yielded the largest loaf is not apparent to the writer, and the loaf size is probably misleading as far as its general character is concerned. The relatively poor showing of B and S duster

bottom flour can doubtless be explained by the lower quality of its gluten, as this particular mill-stream is one of the poorest-quality streams in the mill set-up. These data support the conclusion of Aitken and Geddes (1938) that the great majority of bread flours milled from sound wheats differ mainly in baking strength on account of protein quantity differences.

The durum-wheat-gluten blends showed up remarkably well as compared with the loaf volumes of these flours shown in Table I. The greatest differences in loaf volume in the two sets of data are shown by the stronger flours, while the durum flours gave the smallest differences, and in several instances yielded smaller loaves when baked by themselves than when their dried glutens were blended with the soft-wheat flour. It will also be noticed that the loaves baked from the blend of soft-wheat flour and gluten were, on the average, of better color and texture than the average loaves produced from the flours themselves. This is contrary to the findings of Aitken and Geddes, but may be explained in the present instance by the presence of the durum flours which yield a loaf of distinctly poor color when baked alone. The degrading effect upon color of the durum glutens was masked by the white color of the soft-wheat-base flour in the blends. The same effect is true for the break and duster mill-stream flour glutens which, when baked into loaves with the soft wheat flour, were nearly equal in color to those produced from the spring-wheat-gluten blends. The texture of the blended flour loaves was also superior.

It will be noticed that the average protein content of the original series of flours and that of the blended series were practically identical. The average loaf volumes, on the other hand, were 86 cc. higher for the flours, thus showing that the addition of dried gluten to the soft-wheat flour did not bring the soft-wheat-flour blends to the same level of baking strength as the original flours. This effect may possibly be explained by the presence of poor-quality gluten in the soft winter wheat which lowered the strength of the blend, especially in the case of a superior flour whose gluten quality had now been decreased by the soft wheat gluten contained in the blend. No gluten was washed and blended from flour No. 1, principally because of the lack of a sufficient quantity of this flour for both blending and gluten preparation. The results obtained from flour No. 2, which contained only 1% more protein than flour No. 1, showed no evidence of lower quality in its gluten than the other flours. Comparative loaf-volume results are reported graphically in Figure 3. The gluten-blend loaf volume shown for flour No. 1 is the average value obtained when it was blended with the glutens from all the other flours.

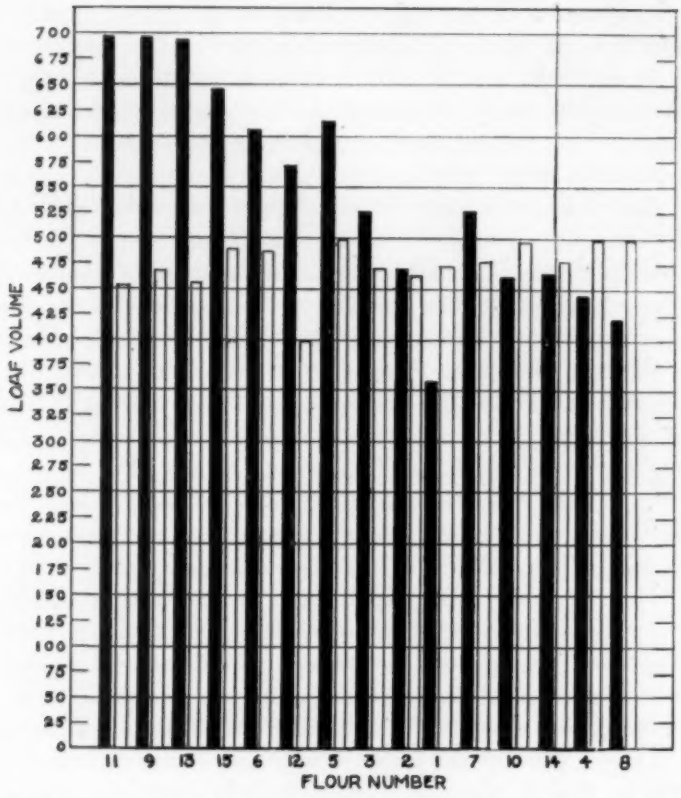

Fig. 3. Loaf volumes of the flours compared with loaf volumes of the dried-gluten and soft-wheat blends. Black bars represent flour loaf volumes; white bars represent gluten-blend loaf volumes.

In Table III are shown the moisture, ash, and protein content of the dried and ground glutens. A substantial range in ash is evident, corresponding roughly to a similar variability in the flour ash. It is evident that a large proportion of the flour ash is carried along with the gluten, although a small percentage of the gluten ash may consist of sodium phosphate absorbed from the washing solution. Some variability in protein content is also noticeable. No relationship between the original flour protein and the protein content of the washed crude gluten is to be noted.

The viscosity and fractionation results obtained on the three series of gluten dispersions are shown in Table IV. The *A* series was prepared from the original wet crude freshly washed gluten. Series *B* was made from wet gluten reformed by the addition of suitable quantities of sodium phosphate solution to the dried gluten. Series *C* was prepared by adding a proportionate quantity of dried gluten directly

TABLE III

MOISTURE, ASH, AND PROTEIN CONTENTS OF THE DRIED GLUTENS

Flour No.	Moisture	Ash [1]	Crude protein[1] (N×5.7)
	%	%	%
2	6.1	0.78	64.3
3	6.7	1.17	63.2
4	6.7	1.06	65.5
5	6.8	0.77	66.9
6	6.3	0.55	68.9
7	7.5	1.38	67.7
8	7.5	1.29	63.2
9	5.9	0.73	67.4
10	7.1	1.38	64.2
11	7.1	1.38	64.2
12	5.6	1.26	65.4
13	5.6	0.82	64.8
14	6.4	1.46	70.0
15	5.3	1.26	65.4

[1] 13.5% moisture basis.

TABLE IV

VISCOSITY AND QUANTITY OF PROTEIN FRACTIONATED FROM THE THREE SERIES OF GLUTEN DISPERSIONS [1]

Flour No.	Viscosity—Centipoises×10²			Protein fractionated per 100 cc. dispersion		
	Series *A*	Series *B*	Series *C*	Series *A*	Series *B*	Series *C*
				mgs	*mgs*	*mgs*
2	200.1	198.7	199.4	632	611	506
3	181.2	179.4	186.0	586	522	506
4	190.2	186.9	183.4	768	739	575
5	211.7	207.8	204.3	739	730	595
6	211.8	198.5	187.9	670	622	335
7	186.2	179.0	175.6	604	645	451
8	185.6	183.8	186.9	566	946	508
9	210.3	204.8	188.4	691	688	410
10	190.9	185.9	185.5	698	686	549
11	210.5	200.9	188.1	659	620	410
12	207.7	199.8	183.0	723	616	513
13	207.2	195.3	205.4	659	570	490
14	192.7	185.0	183.7	534	452	374
15	207.0	195.7	182.9	534	609	489
Means	199.5	193.0	188.6	647.4	646.9	479.4

[1] Adjusted to a concentration of 1400 mg. protein per 100 cc.

to 10% sodium salicylate. The protein concentrations, as pointed out in the table footnote, were adjusted to 1400 mg. per 100 cc. While there is evidence of considerable variation in the corresponding values of series *A* and *B* the quantity of protein fractionated is approximately the same for each series, while for series *C* these results are substantially lower.

These results would lead one to infer that the gluten dispersions prepared from the reformed wet crude gluten more nearly resemble the original wet crude gluten dispersions than would the dispersions prepared from dried gluten without the preliminary addition of water. The mean viscosity results, however, steadily decreased from *A* through *B* to *C*. Series *C* did not resemble series *A* so closely as series *B*. The dried and reformed glutens did not appear to be as extensible and elastic as the undried glutens; they were also somewhat darker in color. They did, however, appear to disperse as readily as the undried glutens in sodium salicylate. This observation is at variance with the observations of Aitken and Geddes (1938), who noted that dried gluten dispersed less in 0.05*N* acetic acid than the corresponding freshly prepared gluten and caused these workers to conclude that the drying process had slightly altered the gluten. Similar alterations had apparently taken place to some extent in the present investigation, although not to the extent of seriously affecting the viscosity and fractionation results of the dispersions prepared from reformed, dried gluten.

When the dried gluten powder was added to the sodium salicylate solution, a distinct difference in properties became evident. The appearance of the dispersions was quite different, flecks of heavily hydrated gluten particles apparently being present in each dispersion. These larger particles were then removed when the dispersion was centrifuged, leaving the more highly dispersed portion of the gluten in the liquid. Some evidence of this is found in the distinctly lower viscosity and fractionation results, indicating the presence of smaller particles as compared with the other two series of dispersions. It is probable that this difference was caused by the action of the sodium salicylate itself upon the dried gluten particles. Table V contains the correlation coefficient calculated from the comparative data presented in Table IV. A high correlation is to be noted between the viscosities of the series *A* and *B* dispersions.

TABLE V

CORRELATION COEFFICIENTS COMPUTED FROM THE VISCOSITY AND FRACTIONATION DATA

Variables correlated		
X	*Y*	r_{xy}
Viscosity, Series *A*	Viscosity, Series *B*	+.9411
Viscosity, Series *A*	Viscosity, Series *C*	+.4618
Fractionation value, Series *A*	Fractionation value, Series *B*	+.2576
Fractionation value, Series *A*	Fractionation value, Series *C*	+.3979

Summary and Conclusions

Glutens were washed from a series of 14 flours which comprised samples milled from hard red spring, hard red winter, soft red winter, and durum wheats. These glutens were dried at a constant temperature, ground into flour-like consistency, and the moisture, ash, and protein contents determined. Calculated additions of these gluten powders were then made to a soft-wheat, low-protein flour to bring the final protein level to the neighborhood of 14%. Bakings were done in triplicate and the results averaged.

The glutens prepared from the hard red spring wheats markedly increased the loaf volumes of the blends, as did one sample of gluten from hard red winter wheat. The soft-winter-wheat gluten was negligible in effect, while the durum-wheat glutens in several instances decreased the loaf volume. The texture and color scores of the blends were superior to the average scores of the loaves from the original flours.

Three series of gluten dispersions were also prepared. Series *A* was from the original wet crude gluten, series *B* from the reformed dried gluten, and series *C* from the dried gluten powder added directly to the sodium salicylate. The resultant dispersions were adjusted to a protein concentration of 1400 mg. per 100 cc. The viscosity was determined and the gluten protein fractionated by suitable additions of $MgSO_4$ solution. The results obtained showed good agreement on the average between series *A* and series *B* as far as the fractionation data were concerned, but series *B* had a lower viscosity on the average than series *A*, probably indicating some alteration in gluten properties. Series *C* had a lower average viscosity and quantity of protein fractionated than series *B*. When compared with series *A* the viscosity differences were still larger, pointing apparently to the presence of smaller gluten particles in series *C*. This conclusion was further borne out by the outward appearance of the three series of dispersions.

In conclusion it may be stated that the method of drying washed crude gluten prepared by Aitken and Geddes (1938) has proved entirely feasible, although there is some evidence of alteration in the properties of the dried glutens. Substantial improvements in loaf volumes were obtained when hard-wheat glutens were added to the soft-wheat flour while the durum glutens tended to lower the loaf volume. Average color and texture scores were improved by the addition of the dried gluten.

Acknowledgments

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THE APPLICATION OF VISCOSITY AND FRACTIONATION MEASUREMENTS TO THE DETERMINATION OF DIFFERENCES IN GLUTENS PREPARED FROM VARIOUS CLASSES OF WHEAT

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Cereal technologists have endeavored from time to time to definitely associate wheat and flour "strength" with the physical and chemical properties of gluten. Such factors as the relative proportions of the gluten proteins present, the viscosity of flour-water suspensions, plasticity and extensibility of doughs and glutens, etc., have been studied by various investigators. As some of these researches were conducted upon doughs and flour-water suspensions which contained large quantities of starch and other non-gluten substances, it seems probable that studies performed with gluten itself as investigational material might yield more pertinent information. Gluten is comparatively easy to prepare, and can be readily dispersed in sodium salicylate solution, as demonstrated by Cook and Alsberg (1931) and Cook and Rose (1934). The properties of the gluten dispersion can then be studied with respect to gluten quality differences. Harris (1937, 1938, 1939) found that the quantity of gluten protein fractionated from sodium salicylate dispersions varied greatly according to the basic loaf volume of the flours from which the gluten had been prepared and the treatment to which the gluten had been subjected while present in the dough prior to washing out.

Harris and Johnson (1939) investigated four flours representative of the hard red spring, hard red winter, soft red winter and durum

wheat classes, using gluten protein dispersions and measuring the quantity of protein precipitated by $MgSO_4$. The flours were mixed into doughs in the customary manner, suitable increments of papain, pancreatin, and yeast water being added. Substantial differences were found in the amounts of protein fractionated among the different classes of wheat flour, and led the authors to believe that hard red spring wheat flour gluten dispersed in sodium salicylate was of larger particle size than that in winter wheat and durum wheat flour gluten dispersions. The results obtained were not quantitative, however, and could therefore not be regarded as highly conclusive.

Harris and Johnson (1940) applied viscosity as well as fractionation methods to the determination of the comparative effects of papain and pepsin upon the fractionation and viscosity values of gluten dispersions prepared from doughs containing these enzymes. Marked differences in effects upon viscosity, rate of flow, etc., were noted between these enzymes, which appeared to indicate differences in size of the protein particle induced by the action of the two enzymes. Differences in the effect of potassium bromate on the two enzymes were also found. It was therefore thought advisable to study further the properties of gluten dispersions prepared from hard red spring, hard red winter, soft red winter, and durum wheat flours, for the purpose of obtaining additional information regarding size of particles in these colloidal solutions. The most desirable procedure appeared to be the use of a series of gluten concentrations to determine the quantity of protein fractionated by $MgSO_4$ and to measure the viscosities of the various dispersions.

Material and Methods

A number of wheats comprising representatives of the hard red spring and winter, soft red winter, and durum classes were included in this investigation. A sample of the hard winter variety, Chiefkan, was also included for comparative purposes as it is generally considered to be distinctly inferior in baking quality. The hard red spring flour was commercially milled but had not been diastated or bleached in the mill. The wheats were milled into straight-grade flour on the Allis-Chalmers experimental mill, and the flours were analyzed for protein and ash content and baked by the malt-phosphate-bromate method. The glutens were washed from these flours by the method described by the senior author in a former publication and dispersed in 10% sodium salicylate solution. Concentrations of 2%, 3%, 4.5%, 6%, 15%, and 20% gluten in sodium salicylate were used. The dispersion of the gluten, which had been divided into approximately

half-gram pieces, was completed in three or four days, during which period the flasks were vigorously shaken by hand. At the end of the dispersion process, the colloidal solutions were centrifuged to remove the undispersed material, and the nitrogen content determined by a modified Kjeldahl-Gunning method. The viscosities of these dispersions were then ascertained by means of the Ostwald pipette at a temperature of 25° C. These solutions were not adjusted to a constant protein concentration for each increment of gluten, but a correction was made from a plotted curve which yielded essentially the same results. The quantity of protein precipitated from the dispersions by the addition of $MgSO_4$ solution to 6% concentration by volume was next determined.

In consideration of results previously obtained by Harris and Johnson (1940) when decided drops in viscosity were obtained with small increases in the pressure under which the dispersions were forced through the pipette, different pressures were also used in determining the viscosities of these dispersions. It was decided to use the Ostwald pipette throughout for this experiment, although when the higher pressures were used with the less viscous dispersions the time of flow was too short to obtain the most precise results. Another slight discrepancy was introduced by the reading of the mercury manometer in the low-pressure regions. In spite of these probable sources of error, however, the values obtained showed very definite trends. A description of the apparatus used in these determinations was published by the authors in a former paper (Harris and Johnson, 1940).

Discussion

In Table I is shown a description of the flours used in this research, with flour protein, flour ash, and data obtained with the use of the malt-phosphate-bromate baking formula. It is evident that a wide

TABLE I

DESCRIPTION, PROTEIN, ASH AND BAKING DATA ON FLOURS USED IN THIS STUDY [1]

Sample No.	Type of flour	Crude protein (N×5.7)	Ash	Absorption	Loaf volume	Color	Texture
		%	%	%	cc.	%	%
1	Hard red spring	13.1	0.51	67	695	94.5	93.0
2	Hard red winter (Turkey)	12.3	0.51	58	616	93.5	92.5
3	Hard red winter (Chiefkan)	10.5	0.48	60	525	92.5	93.5
4	Soft red winter	8.5	0.48	58	469	92.5	94.0
5	Durum (Mindum)	17.1	0.88	63	465	90.0	93.0

[1] 13.5% moisture basis. The malt-phosphate-bromate formula was used in obtaining the baking data.

range of baking strength was included in the samples used in this investigation. These variations were, no doubt, due to differences in protein quality as well as protein quantity. The behavior of the durum sample is especially noticeable as this wheat had a high protein content, but showed poor baking performance, yielding a loaf of low volume and poor appearance. Chiefkan was substantially below the other hard wheats in loaf volume and crumb color.

In Table II are presented the viscosity data, expressed as centi-

TABLE II

DENSITY, VISCOSITY, CONCENTRATION, AND FRACTIONATION DATA FROM SODIUM SALICYLATE DISPERSIONS OF GLUTEN WASHED FROM VARIOUS CLASSES OF WHEAT

Class of wheat	Wet gluten	Density	Viscosity	Dispersed protein per 100 cc.	Protein fractionated per 100 cc.
	%	*g. per cc.*	*centipoises* $\times 10^2$	*mg.*	*mg.*
Hard red spring	2.0	1.0401	155.7	470	198
	3.0	1.0411	169.3	698	304
	4.5	1.0417	192.5	1026	471
	6.0	1.0419	217.5	1361	720
	10.0	1.0430	295.9	2450	1195
	15.0	1.0446	416.5	3300	1767
	20.0	1.0459	594 1	4184	3215
Hard red winter (Turkey)	2.0	1.0408	154.9	469	193
	3.0	1.0410	164.9	653	301
	4.5	1.0416	190.5	978	499
	6.0	1.0419	215.3	1342	692
	10.0	1.0425	279.5	2091	1071
	15.0	1.0441	379.2	3153	1639
	20.0	1.0462	499.2	3956	3247
Hard red winter (Chiefkan)	2.0	1.0347	135.7	425	146
	3.0	1.0365	145.9	656	246
	4.5	1.0372	159.4	864	443
	6.0	1.0370	171.0	1035	587
	10.0	1.0388	221.2	1887	1091
	15.0	1.0401	286.5	2753	1810
	20.0	1.0420	381.3	3238	2437
Soft red winter	2.0	1.0404	151.6	462	158
	3.0	1.0408	157.8	607	237
	4.5	1.0414	176.7	926	418
	6.0	1.0418	198.7	1257	613
	10.0	1.0424	258.5	1986	944
	15.0	1.0438	337.9	2972	1391
	20.0	1.0457	469.6	4030	3013
Durum	2.0	1.0400	146.8	489	160
	3.0	1.0407	155.3	730	260
	4.5	1.0420	172.9	1105	448
	6.0	1.0422	186.4	1376	607
	10.0	1.0421	242.5	2248	885
	15.0	1.0453	336.0	3342	1491
	20.0	1.0462	412.6	4012	3175

poises $\times 10^2$, obtained upon the gluten dispersions of these flours without the application of pressure. The concentrations of dispersed protein as well as the fractionation results are also included. Figure 1 presents these values in graphic form. As the data are rather difficult to evaluate from the table, the discussion will be concerned with the figures. From this figure it is evident that the hard wheats have

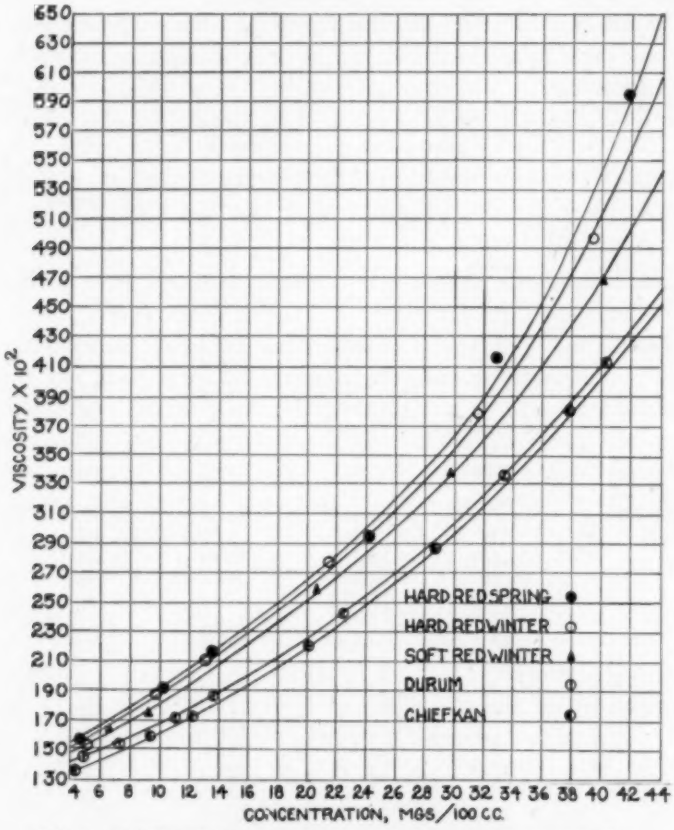

Fig. 1. Relationship between viscosity and gluten protein concentration. Concentration expressed as mg./100 per 100 cc. of dispersion.

higher viscosities than the soft winter or durum wheats throughout the range of protein concentration investigated. It is also evident that the viscosity increases more rapidly than concentration at the higher protein contents, principally because of the adsorption of the dispersion medium to form a hydrated shell around the protein micelle. It would appear that the particles from the stronger wheats were more highly hydrated than the weaker wheats, and this is in agreement with the usual concepts of the hydration capacities of strong and weak

flour gluten. It will be noticed that the hydration of the hard wheats increased more rapidly than that of the soft wheat, and the soft wheat in turn became hydrated more rapidly than durum or Chiefkan.

In Table III the results obtained by computing the hydrodynamic volumes [1] are shown, with the corresponding viscosities. For lyophilic colloids, the quantity ϕ is much larger than the product of the con-

TABLE III

THE HYDRODYNAMIC VOLUMES OF PARTICLES OF GLUTEN WASHED FROM VARIOUS WHEATS AND DISPERSED IN 10% SODIUM SALICYLATE

Concentration—4000 mg. of protein per 100 cc.

Class of wheat	η [1]	η_r [2]	ϕ [3]	Vol. occupied by 1 g. (ϕ/C)
	centipoises $\times 10^2$	*centipoises* $\times 10^2$	%	*cc.*
Hard red spring	544	428.3	28.2	7.05
Hard red winter (Turkey)	570	401.6	27.3	6.82
Hard red winter (Chiefkan)	405	318.9	23.2	5.80
Soft red winter	468	368.5	25.8	6.45
Durum (Mindum)	408	321.3	23.5	5.87

[1] Values interpolated from Figure 1.
[2] $\eta_r = \eta/\eta_0$ where η_0 = viscosity of dispersion medium (127).
[3] Values obtained by interpolation from graph prepared from data presented on page 53 of *Outlines of Biochemistry* by R. A. Gortner, 2nd ed., Chapman and Hall.

centration and the specific volume of the dispersed phase. Similarly, the quotient ϕ/C is much larger than the specific volume. The larger volume may be due to the fact that the dispersed particles are either greatly hydrated or are interlocked and connected to form a micellar structure. As pointed out by McBain (1926), however, true hydration must be a factor in determining the swelling and aggregation and it accordingly appears that high viscosity must be related to the hydration of the dispersed protein particles. The mechanical immobilization of the dispersion medium by the presence of bulky aggregates or by elongated particles that increase the resistance to shear through mutual entanglement must, on the other hand, have a substantial effect upon viscosity. The electro-viscous effect is assumed to be of minor importance in this connection, although its effective magnitude is unknown. From an approximate determination of the migration

[1] These computations were based upon the equation of Kunitz:

$$\eta/\eta_0 = \frac{1 + 0.5\phi}{(1 - \phi)^4}$$

Where η = the coefficient of viscosity of the protein sol.
η_0 = the coefficient of viscosity of the dispersions medium.
ϕ = the percentage of the system occupied by the volume of the disperse phase.
C = concentration of the dispersed phase.

Kunitz, M. An empirical formula for the relation between viscosity of solution and volume of solute. J. Gen. Physiol. 9: 715–725, 1926.

of the dispersed colloid under an electric potential this effect appeared to be relatively small, as would be expected from the pH of the dispersions. The hydrodynamic volumes appear to range themselves in the same order as the "strengths" of the wheats.

The relationships depicted in Figure 2 indicate that plastic flow exists in 40% sucrose, 40% sodium salicylate, hard red spring, hard

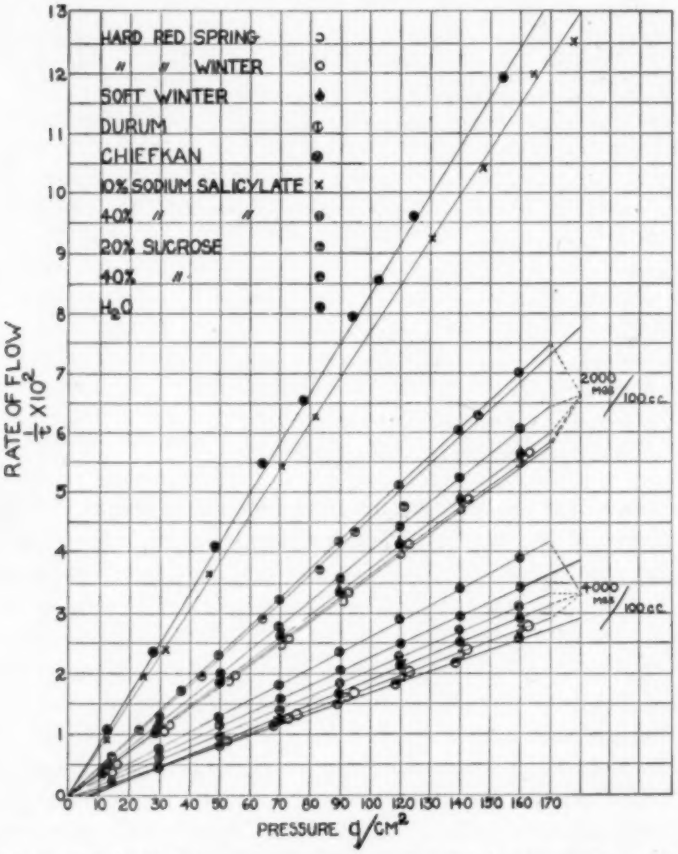
HARD RED SPRING
" " WINTER
SOFT WINTER
DURUM
CHIEFKAN
10% SODIUM SALICYLATE
40% " "
20% SUCROSE
40% "
H_2O
RATE OF FLOW $\frac{1}{t} \times 10^2$
2000 mgs / 100 cc.
4000 mgs / 100 cc.
PRESSURE g/cm²
0 10 20 30 40 50 60 70 80 90 100 120 130 140 150 160 170
0 1 2 3 4 5 6 7 8 9 10 11 12 13

Fig. 2. Relationship between flow and pressure, time (t) being expressed in seconds.

red winter (Turkey), and soft red winter gluten dispersions at concentrations of 4000 mg. of protein per 100 cc. True viscous flow evidently exists in the other liquids investigated, as the lines representing rate of flow with increments of pressure apparently pass through the origin, whereas in the instances mentioned above the lines passed to the right of the point of origin on the pressure axis. A certain amount of "yield" is thus indicated with the application of

pressure before flow actually starts. The plastic flow which apparently exists in the 40% concentrations of sucrose and sodium salicylate must be due to the relatively large number of molecules present. No plastic flow was found by Harris and Johnson in a previous investigation of the relationships between rate of flow and pressure, but the protein concentrations employed in that work were lower than in the present study. The formula used for calculating the total pressure employed in investigating effects of pressure upon viscosity was $[(d_1)(h)] + [(\Delta p)d_2]$, where d_1 is the density of the liquid as determined by a standardized picnometer, h is the average height of liquid in the viscosimeter bulb and corresponds to the point mid-way between the upper and lower constrictions of the bulb, p is the pressure applied, expressed in cm. of mercury, and d_2 is the density of mercury. Where pressure was used the viscosities were calculated from the equation

$$\eta = \frac{[(d_1)(h) + (\Delta p)d_2]t_2}{hd_3t_1}$$

where d_1, h, Δp, and d_2 have the same values as before while d_3 is the density of the reference liquid, t_2 is the time of flow at the corresponding pressure, and t_1 is the time of flow of the reference liquid. Time was expressed in seconds.

The data obtained with pressure were computed to a definite protein concentration by reading off the viscosity values for the desired concentration from the curves shown in Figure 1. This value was then recalculated in terms of flow, $1/t \times 10^2$, and located in Figure 2. By interpolation and suitable calculations employing a proportionality ratio, the corrected value was determined. The viscosity could then be found from the corrected flow-pressure diagram. The additive experimental error inherent in the viscosity-pressure determinations is eliminated by this method of computation.

In Figure 3 the relationships of viscosity to pressure are shown. Two concentrations of sucrose and sodium salicylate solutions were included, in addition to redistilled water and two concentrations of the gluten protein dispersions. Increasing the concentration of the solute or the dispersed phase increased the viscosity of all the liquids examined. Water and 10% sodium salicylate solutions yielded straight-line relationships between viscosity and applied pressure, while 40% sodium salicylate and 20% sucrose solutions showed a small initial decrease in viscosity with increase in pressure. When the sucrose concentration was raised to 40%, however, a marked curvilinear effect was obtained extending to approximately the 90 g./cm.^{2} pressure region. It is thus apparent that this curve in the first por-

tion of the line is related, in the case of true solutions, to the concentration and probably also to the size of molecule, as the sugar molecule carries a hydrate layer which would increase its effective size. From

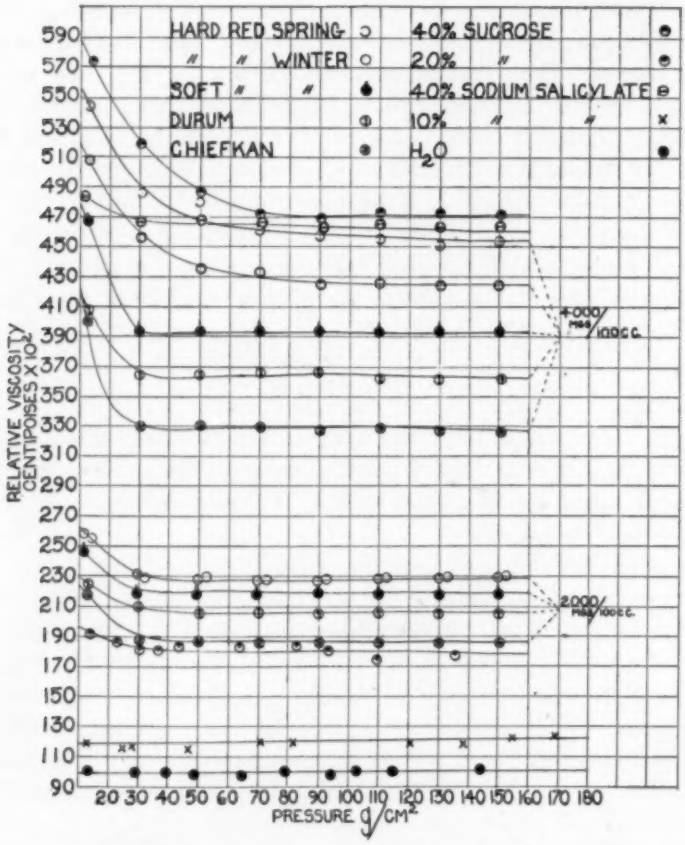

Fig. 3. Relationship between viscosity and pressure, as found in the different solutions and protein dispersions.

an examination of the curves for the gluten dispersions it appears that the same effect of concentration is found as in the instance of the solutions, that is, an increase in the degree of curvature with increase in concentration. This effect was present in the case of each dispersion irrespective of the class of wheat from which the gluten had been prepared, and is probably caused by surface tension, turbulent flow, and resistance at the capillary outflow as pointed out by the authors in a former paper (Harris and Johnson, 1940).

A table of correlation coefficients calculated from a fractionation and viscosity study conducted upon 30 samples of spring wheat flour

are shown in Table IV. A striking positive correlation was found between viscosity and mg. of protein removed from the dispersions by the addition of 6 cc. of concentrated $MgSO_4$ solution to 100 cc. of the gluten protein dispersions. An even higher correlation between these variables was also obtained in the present study. It would therefore be possible to predict with fair accuracy the quantity of protein fractionated from the dispersions from a knowledge of viscosity. As the viscosity determination is the more accurate and precise of the two methods, is more rapid, and obviates the determination of nitrogen by the Kjeldahl procedure, it would appear that viscosity rather than fractionation is the more useful determination. No relationship of any practical significance was found between fractionation or viscosity and baking strength.

TABLE IV

CORRELATION COEFFICIENTS COMPUTED FROM STUDIES CONDUCTED ON FLOURS MILLED FROM NORTH DAKOTA HARD RED SPRING WHEAT—1938 CROP (N=30)

Variables correlated		Correlation coefficient	Probability
X	Y	r_{xy}	P
Protein fractionated (*mg.*)	Viscosity (*centipoises*)	**+.8392** [1]	<.0001
Protein fractionated (*mg.*)	Flour protein (%)	−.0753	>.5530
Protein fractionated (*mg.*)	Loaf volume (*standard*) *cc.*	+.1731	.3453
Protein fractionated (*mg.*)	Loaf volume (*M-P-B*) *cc.* [2]	+.0023	>.5530
Density (*g. per cc.*)	Flour protein (%)	−.2131	.2436
Flour protein (%)	Loaf volume (*M-P-B*) *cc.*	**+.8664**	<.0001
Viscosity (*centipoises*)	Loaf volume (*standard*) *cc.*	+.1373	.4538
Viscosity (*centipoises*)	Loaf volume (*M-P-B*) *cc.*	+.0835	>.5530
FROM DATA IN PRESENT STUDY (N = 35)			
Protein fractionated (*mg.*)	Viscosity (*centipoises*)	**+.9592**	<.0001

[1] Significant values are printed in heavier type.
[2] M-P-B refers to malt-phosphate-bromate baking method.

Summary and Conclusions

A series of gluten dispersions in sodium salicylate was prepared from hard red spring, hard red winter, soft red winter, and durum wheat flours. The effect of protein concentration upon viscosity was determined, as well as the rate of flow in relation to the pressure applied to force the liquid through an Ostwald pipette. The relationship between viscosity and pressure was likewise investigated. The concentrations used varied roughly from 4 to 42 mg. of protein per 100 cc. and the pressure from approximately 12.69 to 160 g. cm. of mercury. The protein concentration of the dispersions was ascertained by the Kjeldahl-Gunning method and the results obtained computed to a definite protein concentration basis.

The results indicated that the size of particle present in these dispersions varied in order of magnitude from hard red spring wheat through Turkey, soft red winter, and durum to Chiefkan. The hydration capacity apparently varied in the same order, which is very close to the general order of water absorption of these respective wheats and is in agreement with the general conception of their baking strengths. Redistilled water, 40% and 20% sucrose, and 40% and 10% sodium salicylate were also investigated. The gluten protein concentrations of dispersions used in the same series of experiments were 2,000 and 4,000 mg. per 100 cc. Large differences in rate of flow were found among these liquids. Redistilled water had the highest and 40% sucrose the lowest rate. Plastic flow was present to some extent in the 40% sucrose and 40% sodium salicylate solution, and in the hard and soft wheat dispersions at the higher concentrations. The durum and Chiefkan dispersions at this concentration were apparently truly viscous, but possibly would show plastic flow at higher concentrations.

An initial inflection in the curve representing viscosity and pressure relationships was obtained in every instance with the exception of redistilled water and 10% sodium salicylate. The degree of this curvature apparently increased with concentration and seemed to indicate a decrease in viscosity with increase in pressure in the lower pressure region. There was some indication that the harder wheats showed a more gradual change at the higher concentrations employed than did the other wheats examined.

The conclusions of Gortner and Doherty (1918) and Sharp and Gortner (1923) regarding differences in the physico-chemical properties of glutens prepared from strong and weak flours, and the relationship of these differences to the colloidal state of the gluten proteins, are confirmed by results obtained in this study. A statement of Rose and Cook (1935) regarding a possible correlation between viscosity values and baking quality is also substantiated to some extent when types of wheat varying widely in "strength" are considered. No significant relationship between these variables existed apparently among the samples of hard red spring wheat examined.

Acknowledgment

The authors wish to acknowledge the assistance of John Monge in preparing the figures and assembling the data contained in this paper.

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A SULPHUR-BEARING CONSTITUENT OF THE PETROLEUM ETHER EXTRACT OF WHEAT FLOUR (PRELIMINARY REPORT) [1]

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(Received for publication February 13, 1940)

It is well known that the baking quality of wheat flour is improved after being extracted by a fat solvent, such as petroleum ether. If the soluble material is recovered and added again to the extracted flour, the bread made from the mixture is usually much poorer than that from either the original or the extracted flour. The emphasis placed by Blish (1936) on the importance of flour lipoids and their possible protein combinations with respect to the quality of wheat bread led us to re-examine petroleum ether extract obtained from fresh patent flour.

An emulsion of the crude lipoidal extract in water was found to give a positive nitroprusside test. This property was lost after exposure to the air, but could be restored thereafter by adding alkaline cyanide. A partial purification of the constituent responsible for this reaction was then attempted. The test was made by mixing a drop of emulsion

[1] Food Research Division Contribution No. 485.

with a drop of 2 *M* KCN on a spot plate. After two minutes a few milligrams of a powdered mixture of sodium nitroprusside and sodium carbonate (1–5) were stirred in.

A petroleum ether extract of flour was concentrated at room temperature and chilled, thereby removing considerable crystalline material that was completely negative when tested for sulfhydryl after a second crystallization.

The evaporated mother liquors could be freed from most of the fat, steroids, and pigments by solution in ether and dialysis against ether through a rubber membrane. The substance sought for was precipitated from the dialyzed ether solution by the addition of four volumes of acetone in the cold.

A method of purification was also devised to avoid the use of acetone and the possible reaction between it and a sulfhydryl group. Instead of dialyzing the ether solution it was mixed with three volumes of 1 *N* HCl in ethyl alcohol, whereupon a precipitate formed. After several hours in the cold, the precipitate was washed three times with absolute alcohol. Thereafter the reactive substance was no longer soluble in ether, so the precipitate was suspended in a large volume of ether for several hours. The material remaining undissolved was collected, dried *in vacuo*, and used for the tests reported here. It appeared to be more concentrated and more stable than that precipitated by acetone. About 0.33 g. was obtained per kilo of flour.[2]

The purified material was insoluble in all the ordinary organic solvents, but soluble in water and in 70% alcohol. The water solution gave a powerful nitroprusside test, but only after reduction with alkaline cyanide. Molisch's test and the biuret reaction were also positive. The solution turned black when boiled with an alkaline lead solution, presumably because of formation of lead sulphide.

On analysis the dried material was found to contain 13.4% of total nitrogen. Protein nitrogen by the trichloracetic acid method (Northrup, 1932) was, however, only 3.9%, and the precipitate formed was gelatinous and unlike that usually obtained with proteins. Sulphur was found to be 2.94%, and phosphorus was absent in a test reliable to 1 part in 2000. When the solution was about two-thirds saturated with ammonium sulphate a precipitate formed that contained the reactive material.

It seems probable that the extractable flour lipoids contain a substance bearing a reversibly oxidizable SH group. The substance is no typical protein, but in some respects resembles a protein derivative. A question of obvious importance is whether it exists originally in

[2] The flour used was freshly milled unbleached patent flour from soft winter wheat. It contained 13.2% moisture, 8.15% protein, and 0.44% ash. We wish to thank the Wilkins-Rogers Milling Co., Inc., of Washington, D. C., for collecting this unbleached flour from the mill stream.

combination with a lipoid or is merely soluble in lipoid-containing fat solvents. This involves the further possibility that the purified material has been chemically changed during its separation from the lipoids. No answer can be given at this time.

The existence of a papain-like enzyme in flour and its influence on the bread-making qualities of the flour are now well known (Balls and Hale, 1938; Jørgensen, 1936). It is possible that the sulfhydryl-containing substance described here may serve as the natural activator of such a proteolytic system, and in this function be capable of modifying the gluten to a marked extent. As yet we have not been able to demonstrate the existence of this substance in commercially bleached flour. If one is willing to assume further that in the original flour the enzyme and such an activator are not always in intimate contact, then the unfavorable influence of added flour lipoids on the quality of the bread is easily explained. In any event we plan to investigate this interesting and reactive material further.

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REPORT OF THE 1938-39 SUBCOMMITTEE ON METHODS OF TESTING SELF-RISING FLOURS

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(Read at the Annual Meeting, May 1939)

Collaborative baking and scoring data were collected on four samples of self-rising flour. Specific directions for baking procedure and scoring were followed. Definitions as given by McKim and Moss (1939) were used for evaluating the scores.

Collaborative Tests

Samples of self-rising flour were sent to each of the collaborators by the chairman. The identity of the flours was kept secret. Definite color scores were specified for flours No. 1 and No. 3, to be used as a

basis for judging the other two flours. The four samples used were the following:

No. 1—A soft wheat family patent, blended from three commercial samples of similar protein and ash.

No. 2—A soft wheat family patent having 0.6% higher protein and color similar to No. 1. (This was a blend of two commercial samples of similar protein.)

No. 3 and No. 4—The same hard wheat cut straight.

The formulae used were: For flours No. 1, 2, and 3, the self-rising formula as given by Walter (1936), (227.7 g. flour, basis 15.0% moisture; 3.4 g. soda; 4.3 g. hydrated mono-calcium phosphate; 4.6 g. salt); and for flour No. 4, additional phosphate over the above formula (227.3 g. flour, basis 15.0% moisture; 3.4 g. soda; 4.67 g. hydrated mono-calcium phosphate; 4.6 g. salt).

With this selection of samples and by using the same procedure and scoring, it was hoped to bring out the following points:

1. The relative biscuit-baking qualities of two soft wheat family patent flours having 0.6% difference in protein.

2. The effect of using excess phosphate on a hard wheat self-rising flour.

3. The ability of several collaborators in various localities to check on the scoring and rating of four *unknown* self-rising flours.

The results are summarized below in Table I.

Analyses, viscosity, and bread-baking tests on the plain flours were made by E. G. Bayfield. As a matter of record, these data are given (by permission) in Table II.

With these data in mind, answers to the three points mentioned above seem to be brought out as follows:

First. The biscuit-baking qualities of the two soft wheat family flours, which differed by 0.6% in protein, were similar, the collaborators scoring them close together in all respects. In Table II it is recorded that both the pH and the viscosity for flour No. 2 were lower than for No. 1. These properties of flour No. 2 may explain why the additional 0.6% of protein over that in flour No. 1 did not make any significant difference in the biscuits. On the other hand, biscuit-baking tests probably are not sufficiently sensitive to detect these comparatively small differences in flours.

Second. The effect of additional phosphate in the self-rising formula of the hard wheat flour did not materially improve the total biscuit score. Two of the collaborators found the color improved one

TABLE I

REPORT OF COLLABORATIVE BAKINGS ON FOUR SELF-RISING FLOURS

	Collaborators					
	A	B	C	D	E	Av.
SAMPLE 1						
pH of biscuit	7.25	7.2	7.2	7.24	7.25	7.23
Oven loss %	16.4	11.3	16.5	8.5	12.2	13.0
Specific volume [1]	2.22	1.70	1.85	1.55	2.01	1.87
Total score	104.4	94	94.2	90	99.6	96.4
SAMPLE 2						
pH of biscuit	7.15	7.15	7.15	7.20	7.20	7.17
Oven loss %	15.0	10.9	16.7	8.3	13.2	12.8
Specific volume [1]	2.13	1.75	1.94	1.55	1.89	1.85
Total score	103.6	95	99	89	98	96.9
SAMPLE 3						
pH of biscuit	7.35	6.75 [2]	7.25	7.41	7.40	7.35
Oven loss %	13.9	10.8	15.1	7.1	12.3	11.8
Specific volume [1]	2.01	2.00	1.78	1.48	1.95	1.84
Total score	89.1	95	78.5	78	89.7	86
SAMPLE 4						
pH of biscuit	7.02	6.65 [2]	7.15	7.08	7.05	7.08
Oven loss %	14.9	10.9	16.2	9.2	12.1	12.7
Specific volume [1]	2.09	1.90	1.78	1.45	1.92	1.83
Total score	91.8	88.0	84.5	77.0	89.8	86.2

[1] Basis weight of dough.
[2] Omitted from average.

TABLE II

REPORT ON THE PLAIN FLOURS USED BY 1938–39 SELF-RISING FLOUR COMMITTEE

	Flour					Viscosity		
Sample	Protein [1]	Ash [1]	Moisture	Absorption [1]	pH 30°	20 g. flour	2 g. protein	No time
	%	%	%	%		° *MacM.*	° *MacM.*	° *MacM.*
No. 1	8.1	.347	9.5	51.9	5.40	70	119	59
No. 2	8.7	.327	10.0	53.2	5.10	63	89	72
No. 3	10.5	.471	9.6	60.0	5.76	142	117	92

	Standard bake				Malt-phosphate-bromate bake			
Sample	Loaf vol.	Grain	Texture	Crumb color score	Loaf vol.	Grain	Texture	Crumb color score
	cc.				*cc.*			
No. 1	552	98	87	100w	520	90	87	100w
No. 2	513	100	94	100w	477	92	91	100w
No. 3	655	98	100	93cw	676	100	100	91cw

[1] Basis 15.0% moisture.

point with the additional phosphate. The pH of the biscuits, as would be expected, was lowered 0.27 of a point.

Third. The total scores assigned to the biscuits did not check very closely. Differences in volume, which are evaluated by actual measurement, were the chief cause of variation between collaborators. The other items on the score sheet, which are evaluated mostly by personal judgment, checked rather closely in most cases. All collaborators agreed quite well in ranking Nos. 1 and 2 close together. They did not agree so well in ranking Nos. 3 and 4. This may be due in part to the fact that two or three of the collaborators were not accustomed to handling hard wheat flour biscuit doughs. Volume differences mentioned above can be caused both by manipulation of the biscuit dough and by the ovens. With the exception of two results by Collaborator B, the pH results on the baked biscuits checked very closely. Part of these pH values were determined colorimetrically and part with the electric pH meter. Oven losses varied considerably between collaborators but averaged out very nearly the same for all four flours. It is interesting to note that the *average specific volumes* of the four samples varied only 0.04 (from 1.87 to 1.83) in spite of the wide differences in the flours.

In Bayfield's data (Table II) the similarity in viscosity of soft wheat flour No. 1 and hard wheat flour No. 3, when based on 2 g. of protein, is quite possibly related to the higher ash content of flour No. 3. In his bread-baking data the crumb-color score and the loaf-volume score seem to correlate with biscuit scores, the color directly and the volume inversely, a large volume suggesting too much strength for best biscuit production when this test formula is used.

Baking Procedure and Score Sheet

The baking procedure recommended follows that reported by Walter (1935) except for the incorporation of the one-fold, two-roll method recommended by the 1937–38 committee, and a slight change in mixing with the Hobart-type mixer, from 10 seconds to 15 seconds after the addition of the liquid.

The values assigned the different items in the score sheet have been retained. For purposes of simplification, it is suggested that the two items "Flour Quality" and "Eating Quality," heretofore listed under "Flavor" for a score of ten points each, be consolidated as one item, "Flavor," valued at 20 points. Under the "Remarks" column anything unusual about the flour or biscuits should be noted. In scoring, the definitions and methods of evaluating given by McKim and Moss (1939) should be followed. In judging color, the use of the

permanent biscuit standards reported by Pearcy and Putnam (1939) may be applied.

It is the consensus of opinion of this committee that since the *biscuit-baking test* is not critical in determining ordinary *variations in flour quality*, it should be considered a test for determining the following properties of a flour or of a self-rising flour mixture: (1) flour soundness and flavor, (2) evaluation of color, (3) proper chemical balance of self-rising ingredients, and (4) leavening power of the self-rising ingredients.

Recommendations

It is the recommendation of this committee that these methods be divorced from soft-wheat testing, and that the program be made more general in character to include all types of both hard and soft wheat flour that may be made into chemically leavened products. It is suggested that the succeeding committee investigate:

1. The possibility of baking to a uniform oven loss as a means of bringing collaborators to a common basis which may result in more uniform biscuit volumes; and
2. The use of standard flours to smooth out personal, machine, and atmospheric differences in performing the biscuit-baking test.

Acknowledgments

Members of this year's committee were: R. A. Barackman, H. V. Moss, Elizabeth McKim, Harold McGhee, C. C. Walker, Elmer Modeer, and O. E. Gookins. Acknowledgment is given for the able assistance of Jay Hedding in Mr. McGhee's laboratory and Claribel Albright in the Quaker Oats Company laboratory.

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1938-1939 REPORT OF THE SUBCOMMITTEE ON TESTING BISCUIT AND CRACKER FLOURS

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(Read at the Annual Meeting, May 1939)

The program followed by the 1938–1939 subcommittee was planned to deal with two of the recommendations made by the previous committee (Brown, 1939), namely: to determine whether there is a correlation between the classification of flours made by the laboratory baking test and that made by a viscosity test after the addition of 1 cc. of lactic acid (as suggested by Triebold), and whether further correlations are possible between laboratory evaluations of cracker flours and shop performance. No work was attempted on cooky flours.

The committee used the same procedure followed by previous committees. In order to secure additional data, four Pacific Coast cracker flours were studied in addition to four from the Middle West. Members of previous committees have agreed satisfactorily on the chemical analyses of the flours used; hence analyses of these flours were made in one Pacific Coast and one Middle Western laboratory respectively.

One member made all the bread-baking tests and sent a loaf from each flour sample to each member of the committee for scoring and classifying for use as sponge and/or dough flour.

Viscosity tests were made following the method outlined by Bayfield (1936), procedures No. 1 and No. 2. On the basis of his own viscosity results and the analytical data furnished with the samples, each committee member classified the flours.

Crackers were baked in commercial shops following schemes used by Reiman (1938) and Brown (1939); *i.e.*, the two sponge flours were baked respectively with the two dough flours from the same district. Because of circumstances beyond control of the committee the proportions used in the bakings on the Pacific Coast were altered. Four barrels of sponge flour No. 9 were used respectively with dough flours No. 11 and 12, while only 3½ barrels of sponge flour No. 10 were used with 1½ barrels of Nos. 11 and 12. These differences may have tended to equalize the results. Two bakings were made on the Pacific Coast flours; three on the Middle Western. Each collaborator scored and evaluated the resulting crackers according to the method outlined in Cereal Chemistry **15**: 37. One member analyzed the baked crackers and another determined the shortometer values.

For the sake of continuity, the flours were designated as Nos. 9, 10, 11, 12, 13, 14, 15, and 16. Numbers 9 to 12 were Pacific Coast flours,

while 13 to 16 were from the Midwest. Table I shows the analytical data and the type of wheat from which each flour was milled. These flours were selected for the work this year on the basis of their analyses and viscosities.

TABLE I

ANALYTICAL RESULTS OF THE FLOUR STUDIED

Sample number	Ash at 15% H_2O	Protein at 15% H_2O	pH value	Used for	Wheat used
9	.403	8.98	5.75	Med. Sponge	Baart & Soft White
10	.416	10.26	5.82	Strong Sponge	Baart & Soft White
11	.372	7.35	5.47	Weak Dough	Soft White
12	.381	7.50	5.73	Strong Dough	Soft White
13	.355	8.40	5.82	Strong Sponge	Red
14	.385	8.28	6.00	Med. Sponge	Red
15	.368	7.83	6.25	Strong Dough	Red
16	.378	7.21	6.10	Weak Dough	Red & White

Discussion of Results

The viscosity results obtained by the collaborators showed quite close agreement with the no-time method. There was a little greater spread among the results with the one-hour digestion method. This may have been due to the slightly greater sensitivity of the method. Table II gives the averages by both methods.

TABLE II

AVERAGE VISCOSITY DETERMINATIONS BY ONE-HOUR AND NO-TIME METHODS

Sample No.	9		10		11		12	
Method	1 hr.	No time	1 hr.	No time	1 hr.	No time	1 hr.	No time
Average	91	64	105	78	47	33	61	41
Sample No.	**13**		**14**		**15**		**16**	
Method	1 hr.	No time	1 hr.	No time	1 hr.	No time	1 hr.	No time
Average	94	73	82	65	68	51	39	29

The results obtained after the addition of one cc. of lactic acid showed a very wide spread between collaborators. The committee members feel that because of this widespread disagreement some study should be made by the Viscosity Committee, or a Section Committee, to see if these results cannot be made to check as well as our final viscosity determinations.

Table III gives the classification of the flours by the collaborators based on the viscosity tests. These classifications are based on the

TABLE III

CLASSIFICATION OF FLOURS ON BASIS OF ANALYTICAL AND VISCOSITY TESTS

Collaborator no.	Flour sample P1 9	P2 10	P3 11	P4 12	M1 13	M2 14	M3 15	M4 16
				ONE-HOUR METHOD				
1	Sponge	Sponge	Dough	Dough	Sponge	Sponge	Dough	Dough
2*	Strong sponge	Very st. sponge	Dough	Dough	Sponge	Sponge	Sponge or Dough	Dough
3	Sponge or Dough	Sponge or Dough	Cooky	Dough	Sponge	Sponge	Dough	Dough
4	Strong sponge	Very st. sponge	Dough	Strong dough	Sponge	Sponge	Weak sponge	Dough
5	Sponge	Strong sponge	Cooky	Cooky	Weak sponge	Dough	Cooky	Weak cooky
6	Sponge	Sponge	Cooky	Dough	Sponge	Sponge	Dough	Cooky
7	Sponge	Sponge	Dough	Dough	Sponge	Sponge	Dough	Dough Cooky
8	Strong sponge	Strong sponge	Weak dough	Weak dough	Sponge	Sponge	Strong dough	Weak dough
				NO-TIME METHOD				
1	Sponge	Sponge	Dough	Dough	Sponge	Sponge	Dough	Dough
2*	Sponge	Sponge	Cooky	Dough	Sponge	Sponge	Sponge dough	Cooky
3	Sponge	Sponge	Cooky	Dough	Sponge	Sponge	Dough	Dough
4	Sponge	Sponge	Dough Cooky	Dough	Sponge	Sponge	Dough	Dough
5	Sponge	Strong sponge	Cooky	Cooky	Weak sponge	Dough	Cooky	Weak dough
6	Sponge	Sponge	Cooky	Dough	Sponge	Sponge	Dough	Cooky
7	Sponge	Sponge	Dough	Dough	Sponge	Sponge	Dough	Dough Cooky
8	Sponge	Sponge	Cooky	Dough	Sponge	Sponge	Strong dough	Wk. do. Cooky

* Classification made from average viscosity tests.

final reading by both the one-hour and the no-time method. The results on the whole are in fairly good agreement by both methods.

If there is any difference, the one-hour method would place the borderline flours slightly higher than they would be placed by the no-time method. Flour No. 10 rates as a sponge flour by the no-time method, but as a very strong sponge by the one-hour method. One collaborator reports, "Almost too strong for a sponge flour."

Flour No. 15 was rated as a dough flour by most collaborators with both methods. However, in two cases it was rated a degree higher.

The classification of flours from the baking test, given in Table IV, on the whole shows close agreement and parallels the classification of the flours by the viscosity test, with the exception of No. 15, which was rated a dough flour by that method but classed as a sponge flour by the baking test. This would tend to confirm Reiman's (1938) observations that "wherever there was a difference in rating of a flour by the two methods, *i.e.*, the baking and viscosity test, the rating of the baking test was a degree higher than by the viscosity test." The classification of these eight flours also seems to confirm Reiman's observation, "that the collaborators were able to reach the same conclusions with

TABLE IV

CLASSIFICATION OF FLOURS BY BAKING TEST

Collaborator no.	Flour sample P1 9	P2 10	P3 11	P4 12	M1 13	M2 14	M3 15	M4 16
1	Sponge	Sponge	Dough	Dough	Sponge	Sponge	Weak sponge	Weak dough
2	Dough or Sponge	Strong sponge	Cooky	Dough	Dough or Sponge	Sponge	Sponge	Cooky
3	Sponge or Dough	Sponge or Dough	Dough	Dough	Sponge	Sponge	Sponge	Dough
4	Sponge	Sponge	Dough	Dough	Strong sponge	Sponge	Sponge	Cooky
5	Sponge	Sponge	Cooky	Cooky	Sponge	Sponge	Sponge	Cooky
6	Sponge	Sponge	Dough	Dough	Sponge	Sponge	Sponge	Cooky
7	Sponge	Sponge	Dough	Dough	Sponge	Sponge	Sponge	Dough or Cooky
8	Sponge	Strong sponge	Weak dough	Dough	Sponge	Sponge	Weak sponge	Dough or Cooky

respect to the flours from loaves baked by someone else as from their own loaves." The results for the cracker scores show the same tendencies as those obtained by the two preceding committees; *i.e.*, "There is a considerable difference between the way individuals score crackers both in valuation and their rating."

Table V shows the rankings of the crackers baked from Pacific Coast flours. The total scores on the two bakes agree only on the No. 4 cracker, which was rated No. 4 on both bakes. On the composite rating of the two bakes, the No. 3 cracker rates third and the No. 4 cracker rates fourth. These crackers were baked with the No. 10

TABLE V

RANKING OF P CRACKERS BY SCORES AND SHORTOMETER VALUES

Collaborator no.	First bake—Sample P1	P2	P3	P4	Second bake—Sample P1	P2	P3	P4
1	4	3	2	1	3	2	1	4
2	3	4	2	1	4	1	3	2
3	2	1	4	2	2	4	1	4
4	1	1	4	2	4	1	2	2
5	2	3	1	4	2	2	1	4
6	1	2	4	3	1	2	4	3
7	3	2	1	4	2	2	1	4
8	1	3	2	4	3	2	4	1
Total	17	19	20	21	21	16	17	24
Rank	1	2	3	4	3	1	2	4

	SUMMARY				SHORTOMETER RANK			
Cracker	P1	P2	P3	P4	P1	P2	P3	P4
First Bake	1	2	3	4	2	1	3	4
Second Bake	3	1	2	4	4	1	3	2
Total	4	3	5	8	6	2	6	6
Rank	2	1	3	4	2	1	2	2

flour as a sponge and rated by both baking test and viscosity test as "sponge" to "very strong sponge," with most collaborators rating it as a strong or very strong sponge by the one-hour digestion method. This flour is probably approaching the upper limits as a sponge flour, and it appears, in this instance, that viscosity by the one-hour digestion method more properly classifies this flour than the no-time method. This would tend to confirm Reiman's (1938) and Brown's (1939) observations, that "the quality of crackers seems to be more dependent upon the type of sponge flour than the type of dough flour used in the formulae."

The shortometer values on the first bake follow the ranking of the crackers, but on the second bake they were not so close. The No. 2 cracker rates the highest by the shortometer and by the cracker scores.

Table VI shows the ranking of the crackers baked from the Midwest flours. The total rankings on the first two bakes were exactly the same. However, in the third bake this was true only for *Cracker C*. As one member of the Committee states, "With all the crackers being first class, it is harder to pick the winner than the loser."

TABLE VI

RANKING OF M CRACKERS BY SCORES AND SHORTOMETER VALUES

Collaborator no.	First bake—Sample				Second bake—Sample				Third bake—Sample			
	A	B	C	D	H	G	F	E	M	N	O	P
1	3	1	2	4	1	1	4	2	1	1	4	3
2	1	3	4	2	3	2	4	1	2	4	3	1
3	4	2	2	1	2	2	1	4	4	2	3	1
4	1	1	4	4	1	3	4	2	3	1	4	2
5	1	4	3	2	2	1	3	4	2	1	4	3
6	4	1	2	3	3	2	4	1	3	2	4	1
7	4	1	1	2	1	2	3	4	4	3	1	2
8	1	3	4	2	2	1	4	3	3	4	2	1
Total	19	16	22	20	15	14	27	21	22	18	25	14
Rank	2	1	4	3	2	1	4	3	3	2	4	1

	SUMMARY				SHORTOMETER RANK			
Cracker	A	B	C	D	A	B	C	D
First Bake	2	1	4	3	2	1	4	3
Second Bake	2	1	4	3	3	2	4	1
Third Bake	3	2	4	1	3	4	2	1
Total	7	4	12	7	9	7	10	5
Rank	2	1	4	2	3	2	4	1

The composite ranking on all three bakes places *Cracker C* in last place and the *Cracker B* in first place, while *A* and *D* rate the same, although on the basis of the first two bakes the *A* cracker would rate second place and *D* third.

Crackers A and *B* were baked with the stronger sponge flour while *C* and *D* were baked with the weaker sponge flour. *Cracker C*, which rates last place, had the very strong borderline No. 15 flour used in the dough, as also did the *A* cracker, which rated second place. Here again the characteristics of the sponge flour No. 13 might be such that it was able to work with both dough flours better than the weaker No. 14 sponge flour.

Again the shortometer values followed the rankings of the cracker for the first bake and were not widely different for the next two bakes. *Cracker C* was rated last by both the collaborators' scores and the shortometer.

The chemical and physical analyses of the crackers (Table VII), while not differing greatly from those reported by the two preceding committees, do show some points of variation. The thickness of ten crackers is about the same as last year, but the count per pound is much lower. Last year, Brown had an average around 140 per pound on all three bakes. This year the count per pound was from 107 to 123, which is about the average found by Reiman's committee two

TABLE VII

CHEMICAL AND PHYSICAL ANALYSIS OF CRACKERS

Bake No.	Cracker No.	Moisture as red'd	Protein N×6.25	Ash	Fat	Shortometer value	Av. pH	Av. thickness of ten	Av. count per lb.
1-P	1-P	5.08	9.58	2.46	11.58	86.6	8.0	2 12/16	122
	2-P	5.20	9.76	2.59	11.85	83.7	8.4	2 13/16	121
	3-P	5.63	10.19	2.17	11.56	87.3	8.0	2 11/16	123
	4-P	5.30	10.19	2.62	12.01	87.6	8.1	2 12/16	121
2-P	1-P	5.20	9.72	2.14	11.72	83.7	7.9	2 13/16	118
	2-P	4.60	9.63	3.17	11.75	67.5	8.2	2 13/16	123
	3-P	5.33	10.15	2.12	11.97	82.7	8.0	3	117
	4-P	4.18	10.41	2.70	11.86	71.9	7.1	2 15/16	120
3	A	4.80	8.75	2.72	13.87	76.0	8.1	2 13/16	115
	B	4.05	8.66	2.70	13.87	70.0	8.1	2 12/16	118
	C	5.25	8.62	2.53	13.34	90.0	8.1	2 13/16	109
	D	3.93	8.58	2.91	13.56	76.1	7.7	2 14/16	111
4	A	5.30	8.36	3.72	14.08	81.6	8.0	2 13/16	114
	B	4.65	8.18	4.14	14.18	80.2	7.8	2 12/16	115
	C	5.45	8.40	3.30	13.60	85.2	8.0	3 1/16	105
	D	5.15	8.09	4.38	13.91	76.0	7.45	2 13/16	107
5	A	4.60	8.76	2.55	13.63	76.6	7.8	2 15/16	108
	B	4.20	8.66	2.15	13.61	80.6	7.6	2 12/16	114
	C	4.60	8.62	2.55	13.47	72.4	8.0	2 14/16	113
	D	3.90	8.49	2.75	13.37	63.9	8.1	2 13/16	114

Chemical analysis by T. E. Hollingshead. Shortometer values by H. J. Loving.

years ago. The average fat content was lower last year than that found this year and two years ago. There seems to be some relationship between the moisture content of the cracker and the shortometer value. The higher the moisture, the higher the shortometer reading.

Conclusions

The work done this year has been essentially a repetition of work done by previous committees. However, it is felt that the results serve as a substantial confirmation of the work of those committees on the evaluation of flours by the baking and viscosity tests.

We find these methods of evaluation hold equally as well with the Pacific Coast flours as with the flours previously studied.

Results seemed to indicate that the one-hour digestion viscosity method gave a sufficiently reliable classification of these flours to warrant further consideration.

This year the viscosity at the one-cc. lactic acid level was observed particularly, in accordance with the recommendation of the previous committee. Discussion of the results observed indicate that at the one-cc. level the system is not sufficiently stable to give a reliable picture.

Recommendations

The Committee recommends:

That further work of this same type be done to build up a greater volume of data to correlate more closely the laboratory evaluation of cracker flours with their actual shop performance.

That further study be made on the scoring of crackers in order to obtain a closer agreement among collaborators.

That a statistical study be made of all pertinent data accumulated by the committees in the last three years; this to be done in order that more practical conclusions may be reached.

Acknowledgments

To the members of this committee, C. C. Armuth, E. F. Evert, H. J. Loving, T. E. Hollingshead, H. O. Triebold, O. P. Skaer, and Miss Pearl Brown, the chairman wishes to express his appreciation and thanks for the hard work and loyal cooperation given.

The chairman also wishes to express his appreciation to the American Cracker Company of Seattle, Washington, whose general manager, R. P. Thymain, and production manager, A. G. Matsten, gave us full cooperation, and to the Kroger Grocery and Baking Co., their purchasing agent, H. H. Wirtz, and their superintendent of cracker bakeries, R. F. Lovell, and to W. S. Culver, who also gave us the best of cooperation.

Also, the chairman is personally indebted to Messrs. Garnatz and Putnam for the many helpful suggestions in planning this work, and to E. G. Bayfield and his staff for baking the bread.

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PRELIMINARY STUDY OF EIGHT TEST METHODS[1]

F. J. COUGHLIN and DONALD WADE

Procter and Gamble Co., Ivorydale, Ohio

(Read at the Annual Meeting, May 1939)

This report is a brief summary of the work done in studying eight methods for testing cake flours preliminary to the collaborative work of the 1938–39 Subcommittee on Methods of Testing Cake Flour. The eight methods, which included the official A.A.C.C. method, were submitted by committee members as being in use to some extent in their individual laboratories.

TABLE I

Formula No.	Flour	Shortening	Sugar	Salt	Baking powder	Milk	Eggs, whole or whites	Type of cake	Mixing method
1	100	37.0	120	3.7	5.5	70.4	56	White layer	Blending (3-stage)
2	100	44.5	111	2.2	4.4	89.0	40	White layer	Creaming
3	100	55.6	142	3.6	6.2	107.0	60	Yellow layer	Blending (3-stage)
4	100	38.0	120	3.2	4.8	80.0	60	White layer	Blending (3-stage)
5	100	57.2	100	2.8	2.1	51.0	64	Yellow loaf	Single stage
6	100	50.0	100	3.0	7.5	75.0	50	Yellow loaf	Single stage
7	100	54.6	140	3.8	5.0	96.0	75	White layer	Blending (3-stage)
8 (Official)	100	25.0	96	1.5	5.8	88.0	32	White loaf	Single stage

The object of the work was chiefly: (1) to examine the methods for sufficiency of detail and experimental directions, and (2) to indicate if possible a formula or formulas which would be satisfactory for future work. This, it was hoped, would guide and expedite the later collaborative work.

Cake Formulas

Table I identifies the test methods which were submitted by committee members. The first eight columns give the actual cake formulas on the common basis of flour as 100. The last two columns show

[1] Report of 1938–39 subcommittee on Methods of Testing Cake Flours.

the type of finished cake produced and the type of mixing employed by the test method.

Flours

Nine samples of flour were submitted by the committee to be used in testing the eight formulas. The analyses [2] of the nine flours are shown in Table II.

TABLE II

	Flour samples								
	A	B	C	D	E	F	G	H	I
Protein (15% m. b.)	7.4	7.6	7.7	7.4	8.3	7.7	8.3	7.8	7.0
Ash (15% m. b.)	0.34	0.32	0.35	0.38	0.37	0.36	0.37	0.33	0.32
pH	5.2	5.1	5.1	5.0	5.0	5.1	5.0	5.1	5.0
Viscosity	46	57	59	27	55	56	52	57	42

Experimental

Eight bakings of nine cakes each were made. Each baking of nine cakes represented one formula with the nine flours. On each of the 72 cakes the following data were obtained: baking loss, volume, symmetry, crust character, tenderness, silkiness of crumb, grain, color, batter temperature, batter pH, specific gravity, and baking temperature and time. Individual photographs of the cakes were made. All of these data, together with specific suggestions for improving details of the formulas, were submitted to the committee chairman.

Discussion

The eight test units were chosen almost at random and varied widely, (1) in relative amounts of ingredients in the formula, (2) in mixing methods, and (3) in the type of cake to be baked (loaf or layer, yellow or white). In addition, the eight methods considered make no attempt to cover all the possibilities for a "best" test method. Thus the work is not broad enough in scope to permit the selection of a best method for differentiating between cake flours as to suitability. However, using this work in part as a guide, the Committee selected one of these test formulas to compare with the Official Method. The comparison was made in later collaborative work which is being reported separately.

[2] These analyses are the means of those obtained in the laboratories of J. W. Montzheimer, H. W. Putnam, and E. G. Bayfield.

REPORT OF THE 1938-39 COMMITTEE ON METHODS OF TESTING SOFT WHEAT [1]

H. W. PUTNAM, *Chairman*

Igleheart Brothers, Incorporated, Evansville, Indiana

(Read at the Annual Meeting, May 1939)

The Committee on Methods of Testing Soft Wheat was set up in 1937 for the purpose of coördinating the activities of three committees of the Association then engaged in work on soft wheat flours. Cereal chemists and their associates in twenty-four laboratories as well as the employees of two commercial cracker bakeries contributed to the work accomplished by the committee in 1938–39.

Complex problems are involved in the proper classification and measurement of soft wheat flours. The ultimate use to which the products are to be put must be kept in mind constantly. Ideal methods of testing these special-purpose flours do not as yet exist. Baking tests must be used. These involve many variables and possibilities of error. To reduce these obstacles to a minimum and to learn how to test the soft flours under conditions which best will reveal their true quality in relation to the ultimate products in which they will be used have been the objectives even though not the achievements of this committee.

The sub-committee on Methods of Testing Cake Flour (J. W. Montzheimer, chairman) made further comparisons between the present test-cake formula and typical commercial-cake formulas. It seems probable that both the lean A.A.C.C. test-cake formula and a richer type may be required to properly classify cake flours. A valuable subproject was executed by Messrs. Coughlin, Wade, and Green. Dr. Stamberg contributed to the standardization of the technique of scoring cakes for grain, symmetry, and volume and prepared photographs to help reduce differences between collaborators. More detailed directions need to be worked out for the procedure itself as well as for interpreting other characteristics of the cakes. In view of improved mixing equipment now available it is probable that specifications for mixing the test batters require reconsideration.

The Sub-committee on Methods of Testing Biscuit and Cracker Flours (Howard M. Simmons, chairman) carried out a project similar to that of its two predecessors but included Pacific Coast flours as well as midwestern flours. This committee has kept the laboratory evaluation of cracker flours closely correlated with shop usefulness. One of the chief problems is a dependable system for scoring the crackers.

[1] General Report.

A thorough study is to be made next year of the data accumulated during the past three years in the hope of discovering trends not apparent before.

The Sub-committee on Methods of Testing Self-Rising Flours (O. E. Gookins, chairman) made some collaborative tests in order further to study scoring suggestions made last year. Whereas, for two years this committee has been part of the Committee on Methods of Testing Soft Wheat, it may be best to separate it since the biscuit baking test is not itself a critical flour test, but rather is a test which frequently is used for determining soundness in flour, flour color, and the balance between the chemical leavening ingredients regardless of whether the flour is soft or hard.

Methods for testing pie flours again were not considered this year.

Definite progress has been made by each sub-committee during the past two years. In view of the increasing interest in soft wheat problems, these co-operative and collaborative efforts should be continued. Definite recommendations concerning soft wheat testing should be prepared as an aid to the committee which may undertake revision of *Cereal Laboratory Methods*.

The author wishes to acknowledge the fine spirit of co-operation which has pervaded the work. Particular recognition and thanks are due the chairmen of the respective sub-committees.

ANNUAL REPORT OF TREASURER

OSCAR SKOVHOLT

January 1, 1940

The membership statement reveals a continuation of the Association's steady growth from the standpoint of numbers. The net increase of 30 members compares with a gain of 36 in the previous year.

The financial statements reveal that the surplus from the year's operations has exceeded that in any previous period. *Cereal Chemistry* income was appreciably increased over that in the previous year with a smaller increase in expenditures. A part of this income was due to a substantial surplus donated to the Association from the 1938 Cincinnati annual convention and assigned by the Executive Committee to the *Cereal Chemistry* fund. General Association expenditures were practically unchanged and there was a moderate increase in receipts.

During the year, the savings account in the Harris Trust and Savings Bank of Chicago was closed and this amount plus a sum from the checking account was invested in U. S. Savings Bonds. This action was authorized by the Executive Committee upon the advice of the Committee on Investments. These bonds, at a cost of $6,000, have a maturity value of $8,000 after ten years. The interest rate is 2.9% compounded semi-annually.

The account with the North Amercian Savings and Loan Association of Missouri still consists of $400 of class A stock, now with interest accrued to a total of $30.89, and $1,600 of class B certificates of undetermined value. Progress is reported in liquidating class B assets through the sale of property. Payments on these properties have not as yet been substantial enough to convert them to class A accounts. It

seems likely that the present book value as placed on this investment may be realized. This Savings and Loan Association has made application for, and has almost been assured of, the acceptance of its accounts by the Federal Savings and Loan Insurance Corporation.

Some of the details of the Treasurer's and Secretary's offices have recently been assumed by an employee of the Association in the office of the Managing Editor. This centralizes the matter of Association membership lists and eliminates considerable duplication of effort.

Sales of *Cereal Laboratory Methods* continue at an essentially unchanged rate with 80 copies being sold during 1939. Only 60 copies of the third edition remain at the end of the year.

DETAILED MEMBERSHIP STATEMENT DECEMBER 31, 1939

	Total	Active	Corporation	Honorary
Membership December 31, 1938	600	545	53	2
New members added during 1939	54	49	5	0
Members reinstated during 1939	7	7	0	0
Members resigned and suspended 1939	*30*	*26*	*4*	0
Members lost by death	*1*	*1*	0	0
Members in good standing, Dec. 31, 1939	630	574	54	2
Net increase in membership, 1939	30	29	1	0

PROFIT AND LOSS STATEMENT

January 1 to December 31, 1939

RECEIPTS			
Cereal Chemistry			
Membership Dues			
Active	$2,016.00		
Sustaining	540.00		
Subscriptions, reprints, back issues and advertising	7,057.01		
1939 accounts receivable	284.14		
Interest on invested funds	110.35		
Cincinnati convention surplus	339.63		
Appropriated by Association	200.00		
Gross receipts		$10,547.13	
Less 1938 income received, 1939		*326.79*	
Net receipts, 1939			$10,220.34
Association			
Membership dues		2,009.00	
Application fees		147.00	
Interest on invested funds		106.60	
Miscellaneous income		1.75	
Net receipts, 1939			2,264.35
Cereal Laboratory Methods sales, 1939		227.45	
Interest on invested funds		17.13	
Net receipts, 1939			244.58
Deceinnal Index			
Received from *Cereal Chemistry*		75.00	
Received from Association		75.00	
Net receipts, 1939			150.00
TOTAL RECEIPTS OF ALL ACCOUNTS, 1939			$12,879.27

DISBURSEMENTS			
Cereal Chemistry			
Cost of printing journal and reprints	$6,190.84		
Net cost of printing		$ 6,190.84	
Cost of editors and miscellaneous services	2,055.68		
Less 1938 account paid 1939	6.34		
Net cost of editing		2,049.34	
Decennial Index—*Cereal Chemistry* assessment		75.00	
Disbursements 1939		8,315.18	
1938 account—uncollectible		6.46	
Total disbursements 1939		$ 8,321.64	
Net surplus 1939			$1,898.70
Association			
Expenses of President's and Vice President's offices and *News Letter*	339.34		
Expenses of Secretary's office	218.08		
Expenses of Treasurer's office	136.05		
Committee expenses	84.52		
Convention ads in *Cereal Chemistry*	20.00		
Kansas City convention report	313.05		
Expenses of officers' section visits	69.45		
Appropriated to *Cereal Chemistry*	200.00		
Decennial Index—Association's assessment	75.00		
Miscellaneous expense	12.10		
Net disbursements 1939		1,467.59	
Surplus 1939			796.76
Cereal Laboratory Methods			
Binding and mailing expenses		68.11	
Surplus 1939			176.47
Decennial Index			
Surplus 1939			150.00
TOTAL DISBURSEMENTS OF ALL ACCOUNTS		$ 9,857.34	
TOTAL SURPLUS			$ 3,021.93

DISTRIBUTION OF NET ASSETS

Cereal Chemistry assets, 1938	$ 5,310.46	
Surplus, 1939	1,898.70	
Assets December 31, 1939		$ 7,209.16
Association assets, 1938	4,544.62	
Surplus, 1939	796.76	
Assets December 31, 1939		5,341.38
Convention Reserve Fund, 1938	1,000.00	
Assets December 31, 1939		1,000.00
Cereal Laboratory Methods Fund, 1938	1,284.16	
Surplus, 1939	176.47	
Assets December 31, 1939		1,460.63
Decennial Index Fund Assets, 1938	150.00	
Surplus, 1939	150.00	
Assets December 31, 1939		300.00
TOTAL ASSETS DECEMBER 31, 1939		$15,311.17

FINANCIAL STATEMENT DECEMBER 31, 1939

ASSETS

Manufacturers Trust Company checking account	$ 4,304.39
Petty Cash Fund, Lincoln, Nebraska	201.62
Emigrant Industrial Savings Bank, New York	868.37
Franklin Savings Bank, New York	532.57
Building & Loan Stock, Kansas City [1]	1,000.00
U. S. Treasury Bonds	2,000.00
U. S. Saving Bonds	6,120.08
1939 income receivable	284.14
GROSS AND NET ASSETS	$15,311.17

NO LIABILITIES

REPORT OF THE AUDITING COMMITTEE

We have examined the books and the report of the Treasurer for the year 1939 and to the best of our knowledge and belief, these are a true and accurate account of the receipts and expenditures of the American Association of Cereal Chemists.

W. R. STOKES, *Chairman*
CHAS. A. GLABAU
R. T. BOHN

CEREAL CHEMISTRY FINANCIAL STATEMENT—1939

JOURNAL INCOME

Membership				
Active 576 @	$ 3.50	$2,016.00		
Sustaining 54 @	10.00	540.00		
				$ 2,556.00
Subscriptions				
Foreign 354	$2,176.69			
Domestic 164	971.90			
		3,148.59		
Advertising	2,245.37			
Accts. receivable	126.30			
		2,371.67		
Reprints	732.94			
Accts. receivable	133.17			
		866.11		
Back issues	603.32			
Accts. receivable	24.67	627.99		
			$7,014.36	
1938 accts. receivable collected 1939			326.79	
				7,341.15

OTHER INCOME

Interest on invested funds	110.35	
Cincinnati Convention surplus	339.63	
Amt. from General Association Fund	200.00	
		649.98
TOTAL INCOME		$10,547.13

[1] Carried on books at value authorized in 1936; now represented by $400 class A stock plus accumulated interest of $30.89 plus $1,600 of class B certificates of undetermined value.

JOURNAL EXPENSE

Journal issues				$ 5,536.08
Reprints				654.76
Salaries				
Editor-in-Chief	$ 499.98			
Assistant Editor	499.98			
Managing Editor	343.16			
Stenographer	390.00			
				1,733.12
Petty Cash				
Petty cash 1938 balance	$199.18			
P.C. reimbursements 1939	325.00			
		$ 524.18		
Less P.C. 1939 balance		201.62		
Gross petty cash expenditures 1939			$ 322.56	
Less 1938 accts. payable paid in 1939			6.34	
Net petty cash expenditures				316.22

OTHER EXPENSE

Decennial Index Fund	75.00
TOTAL EXPENSE	$ 8,315.18
GROSS PROFIT	$ 2,231.95
Less 1938 accounts receivable collected 1939	326.79
Less 1938 accounts uncollectible	6.46
NET PROFIT 1939	$ 1,898.70

We have examined the books of the Managing Editor of *Cereal Chemistry* for the calendar year 1939 and find the same to be correct to the best of our knowledge.

H. H. JOHNSON
C. W. OFELT